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Powdered desiccated liver preparation

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Executive Summary

The utilisation of low value meat cuts and non-meat parts of the animal which make up about 60% of the total carcass weight, is critically important to assist in enhancing the sustainable productivity and profitability of the red meat industry. Bovine livers are rich in nutrients such as iron, zinc, protein and Vitamin A (retinol). These under-utilised co-products have the potential to address the global nutritional deficiencies in developing countries through effectively transforming these raw materials into stable, value-added ingredients to provide a readily accessible and affordable source of complimentary food rich in nutrients. However, the levels of Vitamin A in fresh bovine livers are extremely high and variable, which require a significant reduction to a recommended safe level for intake as routine consumption of large amounts over a period of time can result in toxic symptoms.

This project is part of MLA's initiative to address the micronutrient deficiencies in developing countries and was mainly commissioned for the production of various dried powders (i.e., beef liver, beef meat and placebo) to the required quantity and specifications for nutritional trials in Indonesia. The main focus of the work was to develop a process for the production of powdered dried beef liver that contains Vitamin A at the recommended level safe for intake (i.e., <6,000 µg per 100 g dried powder). Furthermore, the powder quality with respect to the nutritional, functional and microbiological specifications and fitness for human consumption and suitability for export to Indonesia was determined. The development of the manufacturing process, however, was limited to mainly using the standard freeze drying method.

A number of laboratory experiments were conducted to investigate the effects of pre-drying steps (i.e., sample preparation, cooking and hot oil treatment) in terms of producing desiccated beef liver powder with the desired level of Vitamin A. The findings from the laboratory study revealed a significant impact of these processing steps in reducing the Vitamin A level. The results of the laboratory experiments showed that about 89% reduction in Vitamin A can be achieved through a series of processes, such as dicing the raw fresh beef liver into 10-15 mm cubes, subsequently cooking in hot water at 80°C for 15-25 minutes then mincing followed by a double step hot oil treatment at 60°C for 2 hours in each step. The double hot oil treatment was found to be the main processing step to contribute to a major reduction of Vitamin A level. The cooking step contributed in the overall reduction of Vitamin A, in addition to its important role as a thermal treatment of the product for microbiological safety.

A pilot scale manufacturing process was developed based on the processing methods and conditions established in the laboratory study and tested for the production of a larger quantity of dried beef liver powder to the required specifications. The pilot manufacturing process involved a series of food processing steps, consisting of major unit operations such as cooking in hot water, double step hot oil treatment, and freeze drying. The results of the pilot trials showed a Vitamin A reduction of 89% in dried beef liver powder manufactured at this scale with a corresponding 19% production yield of powder. The cooking step imparted a significant weight loss of the product (about 32%) and resulted in a 7% reduction in Vitamin A. The double step hot oil treatment significantly reduced the level of Vitamin A in the dried powder by 78% and also imparted an additional weight loss reduction of 5%. The drying process contributed to the major reduction in weight of the product (i.e., mainly

moisture loss) in the manufacturing process and resulted in a 10% reduction in Vitamin A. Overall, the results of the pilot scale trials in terms of reducing the Vitamin A to the required level in the dried powder are consistent with those obtained in the laboratory experiments, demonstrating the scalability of the process.

The dried beef liver powder contained the required level of Vitamin A. However, there were instances where Vitamin A in the dried liver powder still exceeded the required level due to cases of extremely high levels of Vitamin A in raw fresh beef liver. In these cases, blending with dried meat powder was an ideal option to achieve the required level of Vitamin A as the dried meat powder contained very low amounts of Vitamin A. In addition, it is likely that further degradation of Vitamin A in dried beef liver powder can be achieved in the subsequent storage, depending on drying methods and storage conditions.

Both beef liver and meat powders were found to be good sources of protein and micronutrients (i.e., iron and zinc). In particular, the iron content of the beef liver powder was about 3 times higher than the beef meat powder. However, both micronutrients were significantly affected by the manufacturing process, suggesting that a better understanding of the effect of various processing steps and conditions is also important for the optimal retention of these micronutrients. The placebo powder was also analysed in terms of its nutritional and functional properties and found to contain very little traces of Vitamin A, iron and zinc (as intended) with similar functional properties to the dried beef liver powder and/or beef meat powder.

The pilot manufacturing process for the production of all dried powders was carried out in accordance with a HACCP (hazard analysis critical control point) food safety plan and good manufacturing practices to ensure their fitness for human consumption. The results of the microbiological tests of these dried powders confirmed the compliance of the manufacturing process with food safety requirements. These products were also manufactured to comply with the regulatory requirements for export to Indonesia. A trial shipment of these products to Indonesia, designed to acquire a “pass bill” and to determine the required export documentations, was successful, confirming their suitability for export to this country. Subsequently, a batch of the final products was also successfully shipped to Indonesia. The exportation of the remaining products is currently underway.

In general, this work has generated new knowledge in the manufacturing process of dried beef liver powder with the required level of Vitamin A, and provided invaluable insights to build upon the development and application of the process at industrial scale. However, it should be emphasised that this work was limited to the development and application of a specific drying method (i.e., freeze drying), as the focus was mainly to produce and deliver a powdered dried beef liver that is fit for human consumption and suitable for export. The high costs associated with the freeze drying method preclude the industrial scale utilisation of this drying technology.

The development of a cost-effective manufacturing process for the production of powdered dried products from any raw feedstocks relies greatly on the development and application of the most efficient drying technique. This is due to the fact that drying, the key operation in the process, is an energy-intensive operation and usually affects the microbiological, nutritional and functional qualities of the dried products due to exposure to longer drying times or elevated temperatures. In addition, the selection of the best drying method depends

on the pre/post drying steps that also influence the overall costs of the manufacturing process (e.g., a new drying concept could eliminate the hot oil treatment to achieve the required level of Vitamin A). It should be noted that the selection of the best drying approach is extremely complex due to the diverse factors involved as identified in the literature review, necessitating the need to undertake a detailed techno-economic evaluation of various options to obtain the best drying approach.

It is therefore recommended that further R&D work is undertaken to establish a fundamental understanding and basis to build upon the development of a cost-effective and scalable manufacturing process at industrial scale operations.

- Develop and optimise a new drying concept (e.g., application of ultrasonics) for a cost-effective and scalable process as an alternative to freeze drying to produce dried beef liver powder with reduced Vitamin A levels whilst minimising the impact of the process on micronutrients (i.e., iron and zinc) degradation with better control of the functional attributes (e.g., flowability, solubility) of the dried powder.
- Investigate the effects of pre/post drying processes in terms of process performance (i.e., cost-effectiveness, scalability and effectiveness in reducing Vitamin A levels) and their impact on micronutrients degradation, functional and microbial attributes of the dried product.
- Evaluate the possibility of masking the undesirable flavours of the dried product using microencapsulation and/or alternative masking technologies through improved understanding of the flavour profile as affected by processing using advanced techniques (e.g., GC-MS).
- Assess the potential of capturing Vitamin A in the extraction process as a further co-product (i.e., natural source of Vitamin A) and utilisation of other offals (e.g., heart, kidney, etc) for blending with the beef liver powder to alternatively achieve the required level of Vitamin A in a cost-effective manner.
- Study the stability of Vitamin A and micronutrients at different storage conditions in dried liver powder and in various food product formats fortified with dried liver powder, and develop strategies to minimise the degradation during storage.

The findings from the future work would strengthen the application and adoption of the manufacturing process at industrial scale operations. These would not only assist the red meat processing sector in enhancing their sustainability and profitability (i.e., through capturing more value from under-utilised co-products), but would also provide a readily accessible and affordable source of complimentary nutrient-rich food to address the global nutritional needs of the at-risk children populations (especially in developing countries). It is also expected that the manufacturing process could easily be extended to other meats and meat products.

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1 Background

MLA is undertaking a study to investigate the feasibility of using a powdered desiccated beef liver to prevent micronutrient deficiencies in at-risk Indonesian children aged 12 to 24 months. Provision of complementary foods, rich in micronutrients, could help address deficiencies and ensure optimal growth, development, health and wellbeing in infants and children. The development and implementation of effective complementary feeding strategies is a high priority for the World Health Organisation (WHO) and Food and Agriculture Organisation (FAO).

Several studies, including a study in New Zealand toddlers, partly funded by MLA, have demonstrated the important role red meat plays in preventing micronutrient deficiencies, particularly iron and zinc, and as a result optimising growth and neuro-behavioural development. Liver is recommended by WHO as a complementary nutrient-rich food and powdered desiccated beef liver could provide a readily accessible, culturally acceptable, easily transportable and affordable approach to address micronutrient deficiencies in at-risk populations. The study to be undertaken in at-risk Indonesian children will use powdered desiccated beef liver, administered as a sprinkle on a traditional infant porridge. It will be the first of several trials to develop a safe, efficacious and commercially-feasible product for the target population.

This project mainly covered the development of a process for the production of powdered dried samples (i.e., beef liver, beef meat and food placebo) to the required quantity using the standard freeze drying technique. The quality attributes of the powdered dried samples in terms of the nutritional, functional and microbiological specifications were assessed for their fitness for human consumption and requirements for the preparation of the product suitable for export to New Zealand and Indonesia.

It is well documented that freeze drying is considered to be the best method of drying foods from the product quality point of view. However, the high costs of freeze drying preclude the industrial scale utilisation of this technology. A scoping literature review of alternative drying technologies to the freeze drying was undertaken to identify the most cost-effective drying process for the production of desiccated liver powder.

2 Projective Objectives

2.1 Objective

The main objective of this project was to investigate and develop a process using freeze drying for the manufacture of powdered dried beef liver, beef meat and food placebo samples to the required quantity and determine the qualities with respect to the nutritional, functional and microbiological specifications and document their fitness for human consumption and suitability for export to New Zealand and Indonesia.

2.2 Deliverables

- Delivery of dried beef liver powder (10 kg), beef meat powder (10 kg), beef meat and liver blend powder (10 kg), and placebo powder (10 kg).

- A critical review of the literature on drying of foods which identifies the most cost effective means of drying beef liver to the specifications.
- A report documenting the description of the process by which the powdered desiccated samples (i.e., beef liver, beef meat and food placebo) are produced and the qualities of these dried samples, and describing the estimated value proposition for production of powdered liver by the most cost effective process identified in the literature review.

3 Methodology

3.1 Literature review

A scoping literature review was undertaken to gather baseline information of the existing drying technologies and the associated manufacturing costs (i.e., capital and operating) of such technologies applicable for the production of dried meat powders. Further information of these existing drying technologies was collected in terms of their impact on the nutritional, microbiological and functional qualities of the product. The review was also carried out to identify suitable manufacturers and distributors of the drying technologies currently used in the commercial production of powdered dried liver.

In addition, a comprehensive searches on the US Patent and Trademarks Office (UPSTO), IP Australia, Derwent Innovations Index and the European Patent Office (EPO) websites using the following search terms “Preparation or Process” AND “Desiccated or Dried or Dehydrated” AND “Powdered or Milled or Comminuted” AND “Beef liver or Beef offal” were undertaken to determine for any patents of the process/preparation registered in this space.

3.2 Production of powdered desiccated products

3.2.1 Materials

Samples of Halal certified edible raw fresh beef liver and minced fresh beef meat were obtained from Wagstaff Abattoir Pty Ltd (Cranbourne, Victoria) and Werribee Station Street Meats Pty Ltd (Werribee, Victoria), respectively. A copy of the Halal certificates is attached in Appendix 1. These raw materials were delivered to the CSIRO’s Food Processing facility (Werribee) in a refrigerated Meat Transport Vehicle (MTV). An example of the fresh beef liver centre temperature profiles logged during the refrigerated transport is depicted in Appendix 2.

For the pilot scale trials, chilled raw beef liver samples were immediately sliced into cubes (~10-15 mm) upon receipt (Fig. 1). The slicing process was carried out manually in a refrigerated room (<9°C). Both raw materials (i.e., diced beef liver and minced beef meat) were then stored frozen at -18°C until further processing.

The ingredients used for the production of placebo powder, including food grade maltodextrin (Itochu Australia Ltd), Halal compliance beef flavouring (Sensient Technologies Australia Pty Ltd) and colouring (CHR Hansen Ltd) were obtained from commercial suppliers (Product specifications are attached in Appendix 3). These ingredients were stored at conditions recommended by the suppliers prior to their use.



Fig.1. Manual slicing of fresh beef liver samples for the pilot scale trials.

3.2.2 Methods

Laboratory experiments

A series of laboratory experiments were conducted to develop a processing method for the production of dried liver powder to the desired specifications. The main specification criterion was the production of dried liver powder with the required level of Vitamin A (i.e., <6,000 µg per 100 g dried liver powder). The main focus of the laboratory experiments was to evaluate the processing parameters that are likely to affect the reduction of Vitamin A to the desired level, including sample preparation (i.e., dicing and mincing), and hot oil treatment. Each experiment was carried out using a 100 g of raw beef liver sample. The laboratory hot oil treatments were carried out in 250 mL glass conical flasks incubated in a temperature controlled waterbath to maintain the desired temperature level. Constant stirring was applied to ensure a uniform heating during the treatments.

After a series of screening tests, the effects of hot oil treatment conditions (temperature and time) were further studied. In addition, further experiments were carried out to explore the effect of a double step hot oil treatment utilising the best hot oil treatment conditions. The ratio of raw liver sample and the oil during the hot oil treatment was maintained at 1:2 (w/v) for all the experiments. In all experiments, the samples were initially cooked in hot water at 80°C for 25 minutes prior to the hot oil treatment.

Pilot scale trials

A number of pilot scale trials were conducted to produce the required quantities of various dried powders (i.e., beef liver, beef meat and placebo). The details of the processing methods and conditions for the production of each dried powder are presented below.

Dried beef liver powder

The manufacture of the dried beef liver powder at pilot scale was carried out utilising the processing methods and conditions established in the laboratory experiments. The main processing operations included cooking in hot water, hot oil treatment and freezing drying. Frozen beef liver cube samples were thawed at 5°C cool room for about 24 hours and then

cooked in hot water at 80°C for 15-25 minutes to ensure that the centre temperature of the product reached to the standard conditions for cooking meat and meat products. The centre temperatures of the product were constantly monitored during the cooking process using a hand-held temperature sensor (EuTech Instruments, Singapore). The cooking process was carried in a 300 L heating vessel (Cleveland, Canada) with constant stirring to ensure a uniform heating (Fig. 2). Immediately after cooking, the cooked beef liver samples were then minced in a pilot scale mincer with 11 mm plate (Fig. 3).



Fig. 2. Cooking of beef liver samples with hot water in a 300 L heating vessel.

The cooked/minced beef liver samples were then loaded into a 300 L heating vessel, which was filled with canola oil pre-heated to the desired temperature of 60°C (Fig. 4). The ratio of the product and canola oil during the hot oil treatment was maintained at 1:2 (w/v) and the process was carried out continuously for 2 hours with constant stirring to ensure a uniform heating and aeration. A second hot oil treatment was subsequently carried out under the same treatment conditions (temperature and time) using a new batch of canola oil.

After the second hot oil treatment, the oil was drained in a metal mesh to separate the oil from the solid product. An additional process was carried out to further remove the remaining oil adhering to the product by washing the product with hot water and subsequently centrifuging for 1-2 minutes (Fig. 5). The solids were recovered and placed in plastic bags,

spread in thin layers on metal trays and then stored frozen at -18°C for 2-3 days prior to freeze drying.



Fig. 3. Mincing of the cooked beef liver.



Fig. 4. Hot oil treatment of the cooked/minced beef liver.

Freeze drying of frozen beef liver samples was carried out in a pilot scale freeze dryer (Cuddon FD80, Cuddon Pty, New Zealand) with the capacity of 100 kg of product per batch (Fig. 6). The samples were freeze dried for 4-5 days under the standard freeze drying conditions (Setpoint temperatures at -14°C for primary stage 1 and 26°C for secondary stage 2; Vacuum pressure of 2.8 mbar). The freeze dried samples were then milled in Mauri bowl blender for 2 minutes (Fig. 7) and sieved in 850 microns metal mesh using a SWECO Vibro-Separator (Locker Industries Pty Ltd, Australia) pilot sieving machine (Fig. 8). The bulk powder was then blended using a MANCA ribbon blender (Food Industry Products Pty Ltd, Australia (Fig. 9), packed and sealed in an aluminium foil packaging, and then stored at 5°C cool room until further final filling/sealing into 15-20 g sachets. The final filling/sealing of the dried powders into 15-20 g sachets was undertaken by an external service provider (Australian Vitamin & Sports Nutrition Pty Ltd, Ballina, NSW).

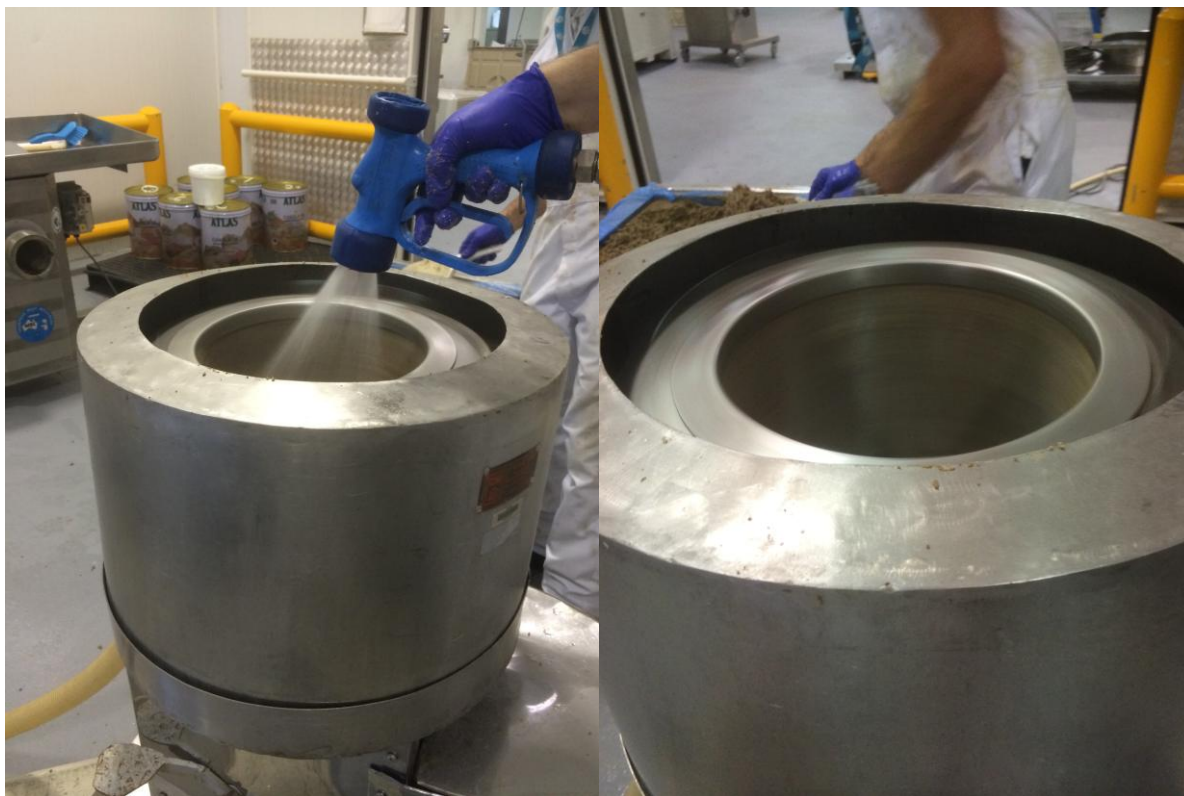


Fig. 5. Hot water washing and centrifuging.



Fig. 6. Photo of the pilot scale freeze dryer.



Fig. 7. Dry milling of freeze dried beef liver.



Fig. 8. Sieving of milled beef liver.



Fig. 9. Blending of dried powders.

Dried beef meat powder

A suite of pilot scale processing trials were also carried out to produce the required quantity of beef meat powder. Frozen minced beef meat samples were thawed at 5°C cool room for 24 hours and then cooked in hot water at 80°C for 15-25 minutes. The cooked meat samples were then washed with hot water and subsequently centrifuge to remove excess fats adhering to the solids. The subsequent downstream processes (i.e., freezing, freeze drying, milling, sieving, blending and packaging) were undertaken using the same processing equipment and conditions as described above for the production of beef liver powder.

Dried placebo powder

A pilot scale processing of dried placebo powder was also carried out to produce the required quantity. The details of the placebo formulation used in the pilot scale trials are presented in Table 1. This formulation was based from the results of the screening tests carried out in the laboratory to obtain the best match of colour, texture and flavour with the meat powdered products.

Table 1. Placebo formulation for a batch of the pilot scale process.

Ingredients	Amounts	
	Percentage (%)	Weight (g)
Maltodextrin Fieldose 30 (Itochu Australia Ltd)	77.21	18,007.3
Malt 205-WA Brown Colouring (CHR Hansen)	3.09	720.8
FruitMax Dark Brown 710WS (CHR Hansen)	1.55	361.8
Beef Flavour XS328 (Sensient)	0.13	31.1
Water	18.02	4,203.0
Total	100.00	23,320.0

The manufacturing process of placebo powder mainly involved mixing of the ingredients by adding maltodextrin first into the mixer/blender and then the liquid ingredients (flavour, colouring and water). The mixing process was carried out using a Mauri bowl blender for 90 seconds. The wet blends were placed in plastic bags and spread thinly on metal trays. The subsequent downstream processes (i.e., freezing, freeze drying, milling, sieving, blending and packaging) were also undertaken using the same processing equipment and conditions as described above for the production of beef liver and meat powders.

3.2.3 Analyses

Duplicate samples of the dried powders (i.e., beef liver, beef meat and placebo) were sent to an external laboratory (National Measurement Institute or NMI, Port Melbourne, Victoria) for standard analyses of Vitamin A, micronutrients (i.e., zinc and iron), proximates (i.e., moisture, fat, protein, ash and carbohydrate), and other heavy metals. In addition, samples of raw beef liver were also sent to NMI for analyses mainly of Vitamin A and moisture. Samples of the dried powders (5 samples per powder) were also sent to Dairy Technical Services Ltd (DTS, Kensington, Victoria) for microbiological testing to ensure their fitness for human consumptions. The details of methods for all of these analyses are attached in Appendices 4 and 5.

4 Results and Discussion

4.1 Literature review

4.1.1 Food drying techniques

Drying is a process applied mainly for the purpose of extending the shelf-life of the food products by reducing their water content (or water activity) to a level low enough to inhibit deteriorative reactions. The removal of water from the food materials during drying can be achieved in different ways, and this variety of methods has led to many drying techniques. Many of these food materials have very diverse physical and chemical properties that need to be dried at different product specifications. The problems of drying are diverse as the intricacies and needs for various materials to be dried at different production scales. There are many different methods of drying food materials, each with their own advantages and disadvantages for particular applications. Over 500 dryer types have been reported in the technical literature, and about 100 types are commercially available (Mujumdar and Law, 2010). This large number of dryer designs is due to the differences in the physical attributes of the product, modes of heat input, operating temperatures and pressures, quality specifications on the dried product, etc. Table 2 summarises a generalised classification of conventional drying methods applied for drying food materials (Sabarez, 2015).

Table 2. A generalised classification of conventional dryers for food materials.

Classification	Types of Dryers (General Characteristics & Applications)
Type of Feed Material	• Particles • Slurry /Paste /Sludge • Liquid Suspension
Processing Mode	• Batch • Continuous
Mode of Heat Transfer	• Convection • Conduction • Electromagnetic (RF, Ohmic, Infrared, Microwave) • Combination (Hybrid)
Energy Sources	• Electricity • Gas (Natural/LPG) • Solar /Wind • Biomass
Mode of Operation	• Cyclic • Intermittent • Continuous
Product Temperature	• Above Freezing Point • Below Freezing Point
Operating Pressure	• Atmospheric • Vacuum • High Pressure

4.1.2 Drying of meat and meat products

Drying is an important critical step in the manufacture of powdered dried meat and meat products (e.g., beef liver). In particular, the production of dried meat powders can be achieved in a number of ways and means. The drying method that can be applied for the production of dried meat powders depends on various factors, including sample preparation (feed type), initial moisture, heat sensitivity, properties of the material to be dried, quality requirements of the product and many others. The most common drying methods that have been reported in the literature for drying of meat and meat products include freeze drying, spray drying, drum/roller drying, hot air drying and fluidised bed drying.

The selection of the drying method for a particular food product is an important step as the drying technique and its operating conditions greatly affect the quality of the dried product as well as its processing costs. In some cases, the choice of the best drying method (i.e., in terms of cost-effective process and maximum product quality) would also significantly depend on pre-drying steps. For example, in the production of powdered dried meat and meat products, the raw solid feedstock can be blended and diluted (i.e., depending on the initial moisture content of the material) to a pumpable slurry, atomised and dried in a spray dryer or dried in drum/roller dryer to produce a powder, or the solid feedstock may be cut into slices or cubes and dried in conventional hot air dryer or fluid bed dryer and then the dried product milled to produce a powder. Often, minor changes in the feed characteristics result in different dryer types being the appropriate choices.

A further challenge in the selection of the best drying method arises from the fact that many food materials have very diverse physical or chemical properties that need to be dried at different scales of production and with very different product quality specifications (Mujumdar and Wu, 2010). Thus, it is especially challenging to establish a universally acceptable rule in the selection of the best drying method, as exemplified by the extreme diversity of factors involved. In most cases, the users must take a detailed assessment for a comparative evaluation of several options for a particular product. Nevertheless, Santivarangkna et al (2007) presented an example on the fixed and operating costs of different drying methods relative to freeze drying (Table 3). It should be noted that the information in Table 3 is specific to a certain product, but could be used as a starting guide for the selection of a cost-effective drying process for meat and meat products. It is however necessary to undertake a detailed techno-economic evaluation of various options to obtain the best drying method (i.e., cost-effective process with maximum product quality).

Table 3. Operational and fixed costs of different drying methods for lactic acid bacteria dehydration (Santivarangkna et al., 2007).

Drying process	Fixed cost (%)	Operation cost (%)
Freeze drying	100.0	100.0
Vacuum drying	52.2	51.6
Spray drying	12.0	20.0
Drum drying	9.3	24.1
Fluidised bed drying	8.8	17.9
Convective hot-air drying	5.3	17.9

Many drying techniques evolved due to the need to produce high quality dried products that are ultra heat-sensitive. Such drying systems include the utilisation of below freezing temperature and under vacuum of the operating pressure (e.g., freeze drying). Freeze drying (also known as lyophilisation) is a drying process in which the food is first frozen then dried by direct sublimation (i.e., phase changes from solid to vapour) of the ice under reduced pressure. It is the choice of drying to guarantee a paramount quality of the final powdered products. However, freeze drying is an expensive process when compared to other drying techniques as it suffers from high production costs, high energy consumptions, and low throughputs (Ratti, 2001). Its cost varies depending on the type of raw materials, the products, the packaging, the capacity of the plant, duration of cycle, etc. (Lorentzen, 1979; Sunderland, 1982). The production cost is approximately eight and four times higher than conventional air drying and spray drying, respectively (Rati, 2001). In particular, the operation costs of an optimised freeze drying cycle could be up to 10 times higher than those for convective hot-air drying (Ratti, 2001). Thus, the high costs associated with freeze drying restrict its usage just to high-value products (i.e., coffee, microorganisms, encapsulated aroma, etc.).

Other drying techniques are based on the type of feed material. For example, for liquid feed, spray drying is still the most common drying method, although rotary drum/roller dryers are also popular (Jangam, 2011). Spray drying is a very expensive technique to use for low value products, mainly because of its low energy efficiency (Jangam, 2011). This method has several advantages, including rapid drying, large throughput and continuous operation. However, due to the relatively high temperatures involved in spray drying processes, this drying technique (spray drying) may cause losses of certain quality and sensory attributes, especially vitamin C, β -carotene, flavours and aroma (Dziezak, 1988).

The majority of dryers used in the food industry are of the convective type; in other words, hot air is used to both supply heat for the evaporation of water and carry away the evaporated moisture from the product. These are by far the most common drying method despite their relatively low thermal efficiency. Hot air produced by indirect heating or direct firing is the most common drying medium. In this type of dryer, the drying medium contacts the material to be dried directly. However, this method of drying requires large amounts of energy and usually imparts significant alterations in product quality and functionality attributes due to the exposure to longer drying times or higher temperatures (Sabarez et al, 2012).

The limitations of convective drying processes may be overcome by combining other novel technologies. In recent years, a number of innovative food processing technologies have been investigated and developed with the aim of improving or replacing conventional processing technologies. These novel or emerging technologies take advantage of other physical phenomena such as sound waves, pressures and electromagnetic fields, which can be applied for the development of new drying concepts for improving the quality of food products through gentle processing. In particular, the application of ultrasonic energy to assist the drying of food materials has been explored for several decades. It has been known for many years that the energy generated by sound pressure waves could enhance a wide range of processes due to a series of mechanisms activated by the ultrasonic energy, such as heat, diffusion, mechanical rupture, chemical effects, and so on (Gallego-Juarez et al., 2007).

A number of investigations have shown the potential of power ultrasound to improve the drying process of various food materials. In particular, a promising approach for the application of ultrasound to assist in the convective food drying of apple slices was developed and tested by Sabarez et al. (2012). This study was carried out to investigate the effect of ultrasound on drying kinetics and product quality attributes using the alternative approach for the application of ultrasonic energy in the convective drying process. The results from this work indicate a significant reduction in drying time (up to 57%) with the simultaneous application of ultrasound on the convective drying of apple slices. This corresponds to a reduction of energy consumption by up to 54% with the ultrasound-assisted convective drying process. In a further study (Beck et al., 2014), the application of a specially designed ultrasonic horn (CSIRO patent) for a completely airborne ultrasound transmission to assist in the convective drying of a model food system was investigated. The airborne ultrasound equipment tested in this work was found to enhance the conventional hot air drying process by significantly reducing the overall drying time (i.e., by more than 60%). This new drying concept offers a promising alternative for a cost-effective drying of meat and meat products. However, further research efforts to optimise the technology for application in various food drying techniques are necessary to provide the basis for developing a new ultrasonic drying technology for adoption in industrial drying practise.

4.1.3 Commercial production of dried beef liver powder

There is very little information on the details of the manufacturing process in the commercial production of dried meat and meat powders (let alone beef liver powder) due to obvious reasons (i.e., commercial competition, intellectual property rights, and so on). However, there are a number of desiccated beef liver powders intended for human consumption that are available in the market (i.e., sold as food supplement). Many of the desiccated beef liver products are also commercially sold as pet food products. These products are dried mainly by freeze drying or spray drying methods. The details of these products (price and quality) can be found at the following websites.

<http://www.aussiewell.com.au/p/8747545/Nutricology-Liver-Powder-Beef-g-Powder.html>

<http://www.nutradry.com.au/products/meat-powders/>

<http://www.drrons.com/organ-and-glandular-supplements/liver-new-zealand.html>

<http://www.directfood.net/?content=product&id=284>

<http://www.toxinless.com/desiccated-liver>

The most relevant commercial drying operation in Australia that is currently supplying dried meat powders in the market is a food manufacturer based in Queensland (Nutradry Pty Ltd, Brisbane). The company has been manufacturing dried meat powders ever since (including beef liver powder). Only recently, the company has stopped producing dried beef liver powder as the market doesn't want to bear the cost of quality drying (pers. comm.). The development of a cost-effective drying system together with the identification of new and further markets for powdered beef liver (e.g., food fortification for Vitamin A, iron and zinc) would therefore re-invigorate the utilisation and value-adding of co-products from the red meat industry, which would also add economic value to the industry.

The company uses a novel drying technique (based on refractance window principle) for the manufacture of various powdered dried food products. The author has been privileged to lead a team of researchers at CSIRO to undertake a detailed “commercial-in-confidence” study of this drying technology to optimise its drying performance at the commercial scale operation (Sabarez and Chessari, 2006). The refractance window (RW) dryer, developed by MCD Technologies, Inc. (Tacoma, Washington, USA), is a drying technique that utilises all the three modes of heat transfer (i.e., convection, conduction and radiation) which occur between the drying medium (water) and the material to be dried for a more energy-efficient drying process. The technology is suitable for producing dried products from liquid and semi-liquid foods (Bolland, 2000). It uses water as a drying medium to transmit heat into the product to be dried. The product is evenly applied to the surface of a conveyor belt system (usually an infrared transparent plastic) that floats on the surface of heated circulating water (Fig. 10). The RW drying technology utilises the refractive principle of the surface of water, which is harnessed by creating a window for the passage of infrared energy.



Fig. 10. Photos of (a) commercial scale RW dryer facility (RWD5 Model, MCD Technologies, USA), (b) wet-feed entry end of the dryer, and (c) dried-product exit end of the dryer (Sabarez and Chessari, 2006).

A number of studies were found in the literature relevant to the RW drying process (Ochoa-Martinez et al., 2012; Caparino et al., 2012; Nindo et al., 2003a; Abonyi et al., 2001; Bolland, 2000; Nindo et al., 2003b, 2004; Clarke, 2004). According to Abonyi et al. (1999), products can be dried in a few minutes with this technology, unlike hot air or tunnel dryers that can take several hours. Nindo et al. (2003a) reported that the drying of pumpkin puree from 80% to 5% moisture content (wet basis) was achieved in less than 5 minutes in both pilot- and commercial-scale RW dryers with a circulation water temperature of 95°C, with a 52% to 70% energy efficiency of the RW drying system.

4.2 Production of powdered desiccated products

4.2.1 Laboratory experiments

Bovine livers are known to contain very high levels of Vitamin A (retinol). This requires a significant reduction to a safe level for intake (i.e., <6,000 µg per 100 g dried powder) recommended by WHO and FAO as routine consumption of large amounts over a period of time can result in toxic symptoms. A number of investigators have studied the stability of Vitamin A (retinol) in solutions and model systems as affected by the presence of heat, oxygen, light, moisture, enzymes, and other oxidising agents (Paquette and Kanaan, 1985; Carvalho et al, 1995; Failloux et al, 2004). However, very little information is available regarding the stability of Vitamin A, particularly in bovine livers during processing and storage. Wilkinson et al (1981) studied the kinetics of Vitamin A degradation in beef liver puree heated at typical canning temperature range (103-127°C) for meat products and observed the rate of degradation followed first order kinetics.

The main focus of the laboratory experiments was to investigate the pre-drying processes that are likely to affect the reduction in Vitamin A in beef liver, particularly by hot oil treatment for leaching Vitamin A into the oil solution. Vitamin A is one of the fat soluble vitamins reported to have good stability during cooking and processing operations, but losses do occur when heated in the presence of oxygen (Lang, 1970; Barratt, 1973). The large number of factors reported in the literature that affected the stability of Vitamin A poses significant challenges in reducing the Vitamin A to the desired level. Vitamin A also appears to be stable for oxidation in oil solution, making the hot oil treatment process ideal for storage and later extraction of Vitamin A from the oil solution.

Cooking is an important pre-drying processing step necessary for pasteurisation (heating <100°C) of meat and meat products in the production of dried powders as the drying process is mainly applied to reduce the amount of moisture (and water activity) in the product to a certain level. This is because the drying process could be carried out at temperatures above or below (depending on drying techniques) of the pasteurisation temperatures recommended for meat and meat products. In the laboratory study, the cooking process of the raw beef liver was carried out at 80°C for 15-25 minutes, depending on the sample preparations (i.e., slicing, mincing and dicing). Under these cooking conditions, the product centre temperatures were found to maintain at 75-80°C for 15 minutes. According to FAO (1992), meat and meat products are considered cooked when the centre of the product is maintained at a temperature of 65-70°C for 10 minutes.

A series of screening tests were carried out in the laboratory to assess the effects of pre-drying steps (i.e., sample preparation, cooking and hot oil treatment) in terms of producing desiccated liver powder with the desired level of Vitamin A. A promising result from these screening tests (raw data not shown) was obtained by dicing the raw beef liver into cubes (~10-15 mm), subsequently cooking at 80°C for 25 minutes and then mincing followed by hot oil treatment. Based from the results of the screening tests, further experiments were undertaken to specifically investigate the effect of hot oil treatment conditions. Table 4 shows the levels of Vitamin A in the dried liver powder as affected by hot oil treatment conditions. It should be emphasised that the amounts of Vitamin A presented in Table 4 are based on a 100 g of sample with varying amounts of moisture. This artificially concentrates the Vitamin A and masks the loss caused by the treatment. In order to make a meaningful comparison,

the % reduction in Vitamin A was calculated based on the dry matter content of the product to correct the effect of the amount of water in the product.

It can be seen from the table that the highest reduction in Vitamin A (about 83.7%) was achieved with hot oil treatment at 60°C for 2 hours. Under these conditions, the level of Vitamin A of the dried powder was about 6,100 µg per 100 g of dried powder (with 2.2% moisture). This was achieved by utilising a batch of raw beef liver samples with Vitamin A of 12,000 µg per 100 g of raw beef liver (with 68.6% moisture). However, the Vitamin A level of the dried powder was a little bit over to the desired level of 6,000 µg per 100 g of dried powder.

Table 4. Effect of sample preparation and hot oil treatment on Vitamin A reduction in freeze dried beef liver powder (*All treated samples were initially cooked in hot water at 80°C for 25 minutes prior to hot oil treatment*).

Treatment Description	Moisture (%)	Vitamin A	
		(µg/100 g sample)	(% reduction)
Raw fresh beef liver	68.6	12,000	-
Hot oil treatment (60°C for 1 hr); diced sample	2.3	8,700	76.7
Hot oil treatment (60°C for 2 hr); diced sample	2.2	6,100	83.7
Hot oil treatment (60°C for 2 hr); minced sample	2.0	18,000	51.9
Hot oil treatment (80°C for 15 min); diced sample	3.4	20,000	45.8
Hot oil treatment (80°C for 30 min); diced sample	2.3	10,000	73.2
Hot oil treatment (80°C for 30 min); minced sample	2.3	11,000	70.5

Note: The % reduction in Vitamin A (as compared from raw beef liver) are calculated based on the dry matter basis to take into account the amount of water in the product.

Further experiments were then carried out to investigate the effect of a double step hot oil treatment in reducing the Vitamin A level in beef liver. This was undertaken by sequentially treating the product twice with hot oil under the same conditions using a new batch of oil for each step. Table 5 shows that the subsequent second hot oil treatment resulted in a further 9% reduction in Vitamin A. The results also show that the overall reduction in Vitamin A that can be achieved is up to 89% under these conditions. However, as can be seen from this table the Vitamin A level of the dried liver powder (which is about 8,100 µg per 100 g of dried powder) still exceeded to the required level of Vitamin A.

Table 5. Effect of double step hot oil treatment on Vitamin A reduction in freeze dried beef liver powder (*All samples were diced and initially cooked in hot water at 80°C for 25 minutes prior to hot oil treatment*).

Treatment Description	Moisture (%)	Vitamin A	
		(µg/100 g sample)	(% reduction)
Raw fresh beef liver	69.3	23,000	-
Hot oil treatment (60°C for 2 hr)	1.5	15,000	79.7
Double step hot oil treatment (60°C for 2 hr)	1.4	8,100	89.0

Note: The % reduction in Vitamin A (as compared from raw beef liver) are calculated based on the dry matter basis to take into account the amount of water in the product.

The result is consistent with the fact that these experiments were carried out utilising a different batch of raw beef liver samples, which contain almost twice the amount of Vitamin A than in the previous batch. The Vitamin A in this batch of raw beef liver was about 23,000 µg per 100 g of raw beef liver (with 69.3% moisture). In view of these, a further analysis on the variability of Vitamin A levels in raw fresh beef liver was conducted. The findings from this

analysis as shown in Table 6 revealed a significant variation in the levels of Vitamin A between liver samples from individual animal. It appears that the variation in Vitamin A in raw fresh beef liver was extremely significant (i.e., one individual beef liver contains over 3 times Vitamin A than others). This is quite a challenge from the processing perspective in achieving the required level of Vitamin A.

Table 6. Vitamin A content of individual raw fresh liver samples (*average moisture content of 69.3%*).

Liver No	Weight of individual liver sample (kg)	Vitamin A of individual liver sample ($\mu\text{g}/100\text{ g sample}$)
1	3.368	15,000
2	5.340	38,000
3	3.275	21,000
4	4.220	14,000
5	4.420	25,000
6	3.927	13,000
7	3.992	33,000
8	3.677	16,000
9	5.209	43,000
10	4.480	28,000

A possible solution is to blend the dried beef liver powder with beef meat powder in order to achieve the desired level of Vitamin A. Beef meat is known to contain low levels of Vitamin A and can similarly be processed into dried powder using low value meat cuts (e.g., trimmings, etc). A preliminary laboratory freeze drying experiment using minced beef meat shows that the resulting beef meat powder only contains about 83 μg of Vitamin A per 100 g of dried meat powder (with 1.3% moisture). In addition, it is likely that a further degradation of Vitamin A in dried beef liver powder can be achieved in the subsequent storage of the dried powder. However, the extent of such degradation is dependent on many other factors, including the storage temperature, exposure to light and oxygen (i.e., type of packaging), and so on. Moreover, it should be noted that this project is focused in transforming the raw beef liver into desiccated powder by mainly using the standard freeze drying method in order to achieve the required quantity and product specifications. As the process of drying usually involves heating and exposure to hot air, it is likely that further reduction in Vitamin A level can be achieved by drying, depending on the drying methods and conditions. This warrants a future investigation of the impact of the drying process not only to achieve the desired level of Vitamin A with better control, but also to obtain a cost-effective and commercially scalable drying process.

4.2.2 Pilot scale trials

Process development

The project is mainly focused on the development of a process specifically for the production of dried beef liver powder with Vitamin A to the required level. The manufacture at a larger scale is necessary to effectively produce the required quantity and to demonstrate the scalability of the process. Fig. 11 shows the details of the pilot scale manufacturing process developed for the production of dried beef liver powder utilising the processing methods and conditions established in the laboratory experiments and through a series of pilot scale trials.

The manufacturing process involved a series of food processing steps, consisting of major unit operations such as cooking in hot water, double step hot oil treatment, and freeze drying. The results from comprehensive searches on patents suggest that there is no evidence of any registered patents that may be infringed with the manufacturing process developed in this work, although there are a number of patents surrounding other various dried beef liver preparations (Wang, 2010; Lu, 2007; Procter, 1978).

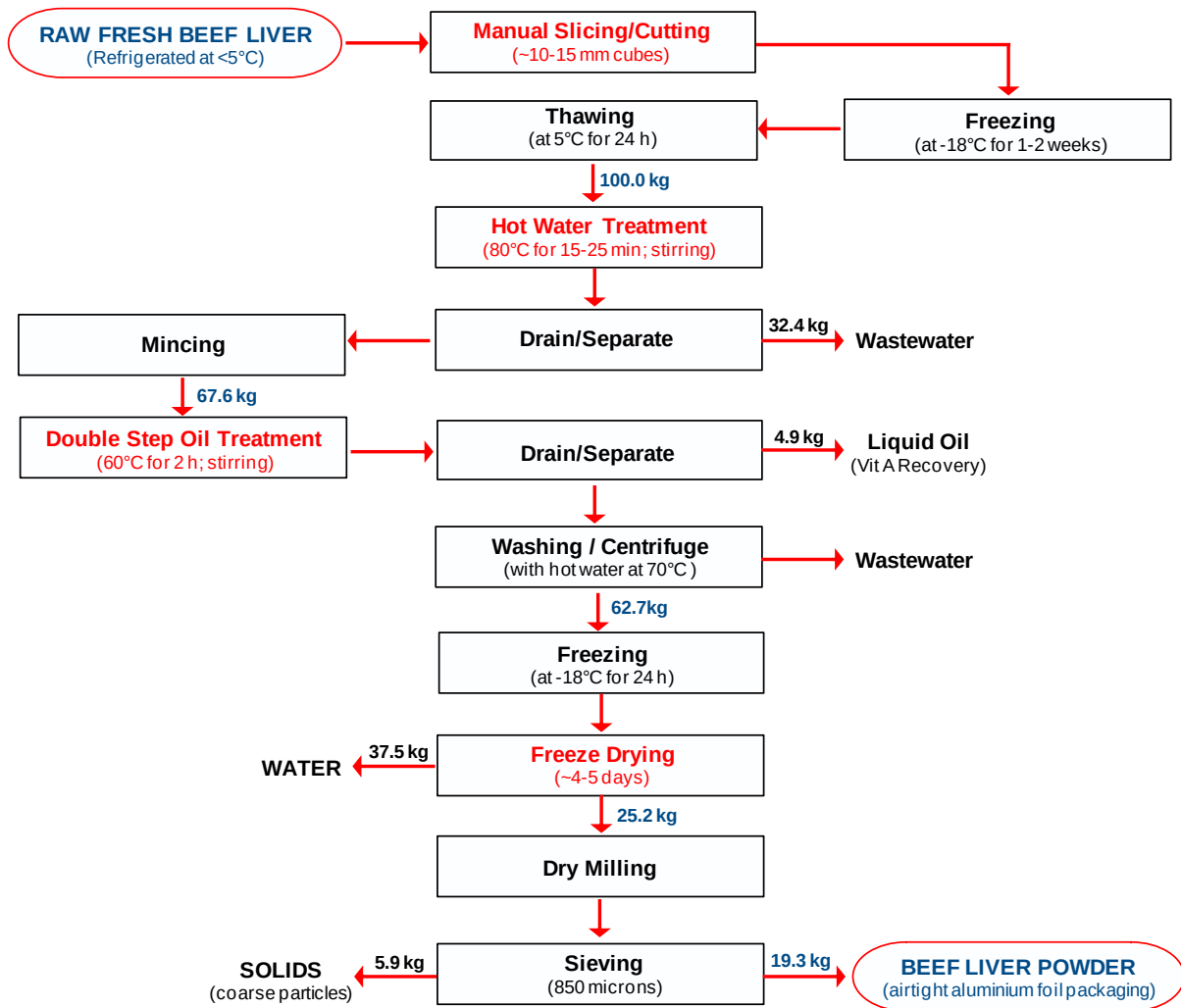


Fig. 11. A schematic diagram of the pilot scale manufacturing process for the production of dried beef liver powder.

A typical batch of the pilot scale process can produce about 19.3 kg of dried powder (with 1.5% moisture) from a 100 kg feedstock of raw fresh beef liver (with 70.3% moisture). The details of the production yield recovery at various steps in the manufacturing process are also presented in Fig. 11. Table 7 presents the production performance and the corresponding % reduction in Vitamin A level after each major step in the pilot scale manufacturing process for the production of dried liver powder. The results show about 89% reduction in Vitamin A of the dried beef liver powder manufactured at the pilot scale (i.e.,

1000 times larger than the laboratory scale). This is consistent with those found in the laboratory experiments, demonstrating the scalability of the process.

Table 7. Effect of double step hot oil treatment on Vitamin A reduction in freeze dried beef liver powder at pilot scale (*All samples were diced and initially cooked in hot water at 80°C for 25 minutes prior to hot oil treatment*).

Parameters (major processing steps)	Production		Moisture (%)	Vitamin A (% reduction)
	Weight (kg)	% Weight loss		
Raw fresh beef liver samples (diced)	100.00	-	70.3	-
After cooking and then mincing	67.65	32.35	59.0	7.5
After double step hot oil treatment*	62.75	37.25	54.5	78.0
After freeze drying	25.17	74.83	1.4	88.3
After grinding and sieving	19.26	80.74	1.5	89.2

Note: * This includes subsequent washing with hot water and centrifuge.

In particular, the results in Table 7 indicate that the cooking step imparted a significant weight loss of the product (about 32%), which could be mainly due to the removal of water from the product. This is in accordance with the weight reduction reported in the literature during cooking of meat and meat products (FAO, 1992). In the production of powdered meat and meat products, cooking is an important processing step prior to drying to reduce or eliminate the microbial content in the product, especially if the drying process is carried out at low temperatures (e.g., freeze drying). The cooking process can vary considerably in treatment conditions (i.e., temperature and time) depending on the type of product. Thus a better control of these conditions is important to achieve a compromise between microbiological safety and product quality requirements. The cooking treatment should be intensive enough to accomplish adequate microbial reduction whilst keeping it to a level just high enough to prevent deterioration of product quality. For meat and meat products, cooking is usually carried out in the temperature range of 60-85°C (FAO, 1992). On the other hand, the results also show a 7% reduction in Vitamin A attributed by the cooking step. This could be due to combined effect of heating and exposure to oxygen (constant stirring) during cooking.

Table 7 also shows a further reduction in weight of the product (about 5%) as affected by the double step hot oil treatment. The observed modest weight reduction during double step hot oil treatment could be mainly due to some losses of the solids (i.e., fine particles drained with the oil solution). It should be noted that the cooked liver samples were minced prior to hot oil treatment to reduce the particle size of the solids to enhance the leaching of Vitamin A into the oil solution. The hot oil treated materials were then washed with hot water and centrifuge to further remove excess fats adhering to the solids. These could have further contributed to the observed weight reduction of the product. A thorough washing with hot water of the hot oil treated solids is necessary to obtain a free-flowing powder. Without this step, the subsequent drying process would be very slow and difficult and the dried product after drying would be very sticky due to the excess fats (Fig. 12).

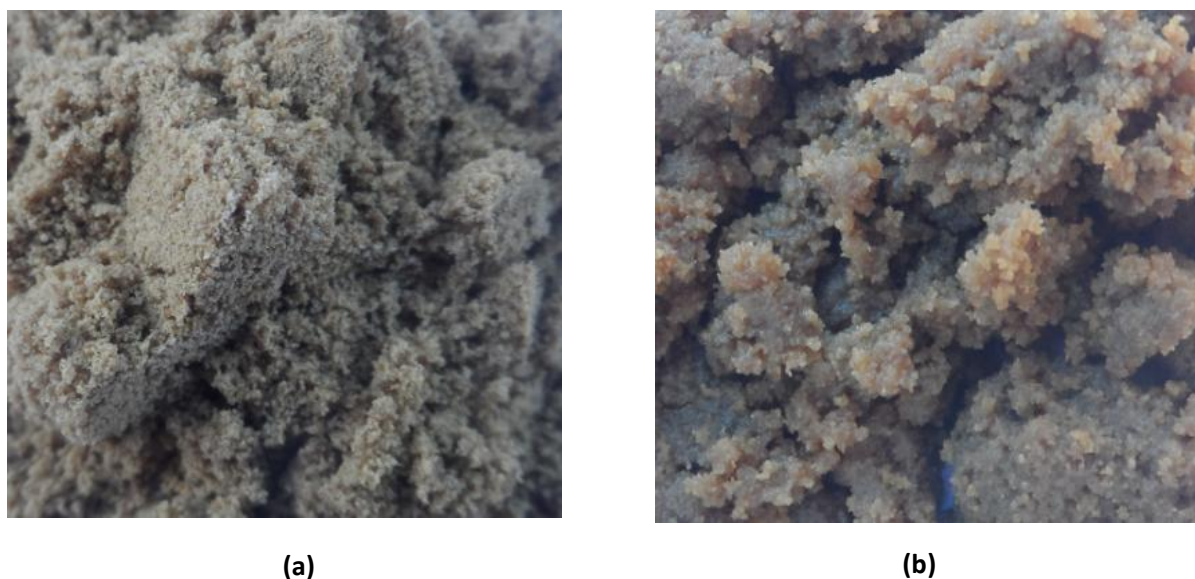


Fig. 12. Photos of (a) dried liver powder (b) wet dried liver powder

It should also be borne in mind that the double hot oil treatment was designed as the main step in the manufacturing process for the reduction of Vitamin A in the solids. Vitamin A is a fat soluble component in foods and was found in the initial laboratory experiments to readily leach into a vegetable oil solution, depending on treatment conditions. This observation is also confirmed from the results in the pilot scale trials as demonstrated in Table 7. As can be seen in the table, the double step hot oil treatment significantly reduced the level of Vitamin A in the processed solids by 78% from the Vitamin A content in the raw fresh beef liver samples. The leaching of Vitamin A into the oil solution can be visually observed on the changes of the colour of the vegetable oil before and after treatment (Fig. 13). Vitamin A can then be extracted from the oil solution for further use. In addition, the hot oil treatment could have resulted in losses (or changes) in flavours of the product as many of the flavour compounds are oil soluble and volatile at higher temperatures. However, a detailed study in this area is required to characterise the impact of the process on flavour profile.

Drying is the main step in the manufacturing process for the production of powdered meat and meat products. It is a processing step commonly used to reduce the amount of moisture (hence water activity) in food products to a level safe for preservation. In the current work, the drying process was mainly limited of using a standard freeze drying technique to achieve the desired product specifications. This was undertaken by using the standard conditions typical in freeze drying of food products (i.e., no detailed study of the drying process was undertaken in the current phase of the project as it was proposed to be undertaken in the next phase of the project). Obviously, the drying process has contributed the main reduction in weight of the product during the manufacturing process as shown in Table 7. It was found that the product losses weight by 75% from its original weight (or a further reduction in weight by 38% after the double step hot oil treatment).

The observed weight loss during drying is consistent with the removal of water from the product, as demonstrated from the significant reduction in the measured amount of moisture in the product. However, the overall weight loss of the product after freeze drying was found

to be greater than the initial water content in the raw materials, indicative of some solid losses during the previous steps (i.e., cooking, hot oil treatment and so on) prior to freeze drying. In addition, freeze drying was also observed to contribute to a further reduction in Vitamin A (about 10%). This could be due to the exposure of the product to oxygen as air is constantly circulated during the drying process, although the drying process was carried out at very low temperatures. This suggests that the drying process (i.e., depending on drying techniques and conditions) could play a further significant role in achieving the desired level of Vitamin A in beef liver powder.

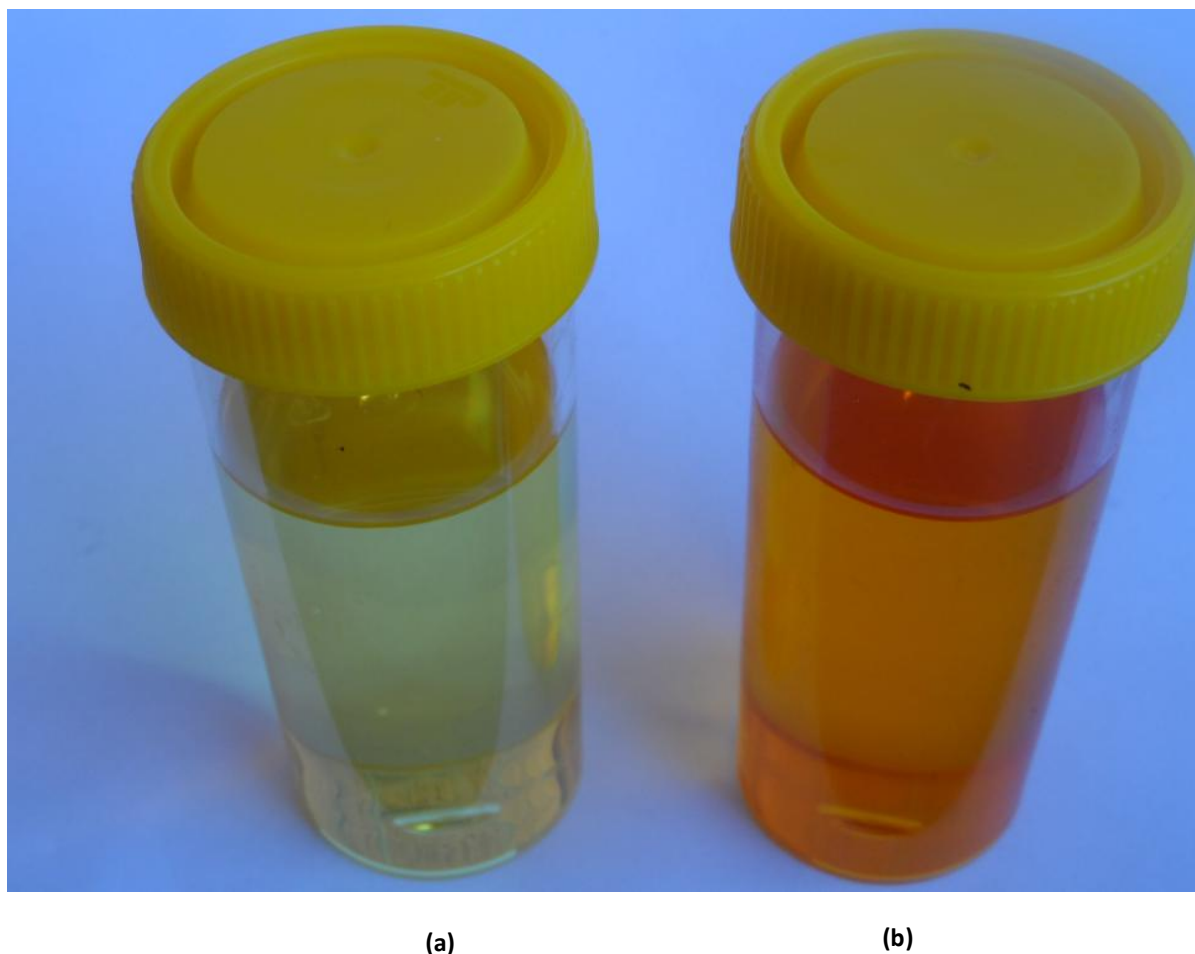


Fig. 13. Photos of (a) fresh canola oil and (b) used canola oil after hot oil treatment with beef liver samples.

Product quality assessment

The final dried powders were assessed in terms of their nutritional, functional and microbiological qualities. These powders (particularly the beef liver powder) were evaluated with respect to the required levels of Vitamin A (i.e., <6,000 µg per 100 g of dried powder). The results presented in Table 8 show that the developed pilot scale process can produce dried beef liver powder with Vitamin A content less than 6,000 µg per 100 g of dried powder. However, this is highly dependent on the Vitamin A level of feedstock (i.e., raw fresh beef

liver) used in the manufacturing process. In a particular batch, the average Vitamin A content of the raw beef liver samples was about 15,000 µg per 100 g of raw liver (with 70.9% moisture) (Table 9). This means that when the initial Vitamin A levels in raw fresh beef liver are very high (e.g., >18,000 µg per 100 g of raw liver), the pilot scale process is not sufficient to produce a dried liver powder with Vitamin A to the required level as observed in another batch of pilot scale trial (data not shown). In this case, blending with dried meat powder is an ideal option to achieve the required level of Vitamin A as the dried meat powder is shown to contain very low amounts of Vitamin A, i.e., 22 µg per 100 g of dried powder (Table 8).

Table 8. Nutritional profiles of the final dried powders.

Nutritional specifications	Dried products			
	Beef liver powder	Beef meat powder	Beef meat & liver blend powder	Placebo powder
<u>Proximates:</u>				
Moisture (%)	1.5	1.4	2.6	2.6
Fat (%)	21.5	22.4	19.2	<0.2
Protein (%)	62.3	65.9	71.2	0.3
Ash (%)	3.5	1.6	7.1	<0.1
Carbohydrate (%)	11	9	<1.0	97.0
<u>Vitamins:</u>				
Vitamin A (µg/100 g sample)	4250	22	175	<5.0
<u>Trace Elements:</u>				
Iron (mg/kg)	160	45	61.5	<2
Zinc (mg/kg)	150	180	155	0.265
Antimony (mg/kg)	<0.01	<0.01	<0.01	<0.01
Arsenic (mg/kg)	0.02	0.032	0.028	<0.01
Cadmium (mg/kg)	0.087	<0.01	0.018	<0.01
Copper (mg/kg)	61.0	2.6	15.0	0.089
Lead (mg/kg)	0.07	0.013	0.026	0.017
Mercury (mg/kg)	<0.01	<0.01	<0.01	<0.01
Selenium (mg/kg)	0.48	0.2	0.28	<0.01
Tin (mg/kg)	0.27	0.024	0.048	<0.01

Note: Blend ratio of liver powder to meat powder is 20:80.

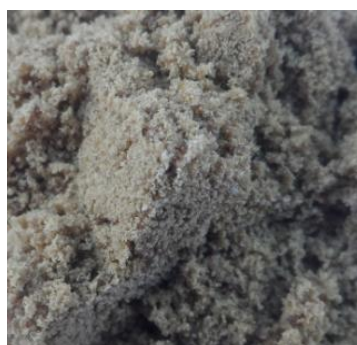
All the final dried powders were also tested with their nutritional profile, including proximate compositions (i.e., fat, protein, carbohydrate, ash and moisture), micronutrients (i.e., zinc and iron) and other trace elements normally performed for foods. The results of these analyses are also presented in Table 8. The results confirmed that both beef liver and beef meat powders are equally a good source of protein. The iron content of beef liver powder was much higher (3 times more) compared with the beef meat powder. The beef meat powder was found to contain a little more of zinc than the beef liver powder. In general, both micronutrients (iron and zinc) were found to be significantly affected by the manufacturing process (Table 9). For example, the iron content in dried beef liver powder was found to be 46.5% less than in raw fresh beef liver, while the zinc content of the dried beef liver powder was 52.7% less than in raw fresh beef liver. This suggests that both micronutrients are sensitive to the process and detailed understanding of the effect of various processing steps and conditions in the production of dried powders are also important in achieving the optimal levels of these important micronutrients.

Table 9. Nutritional profiles of a beef liver powder processed at a pilot scale.

Products	Moisture (%)	Vitamin A (µg/100 g sample)	Iron (mg/kg)	Zinc (mg/kg)
Raw fresh beef liver	70.95	15,000	83	83
Dried beef liver powder	10.1	4,700	130	115
% Reduction	-	89.8	46.5	52.7

Note: The % reduction in Vitamin A, iron and zinc (as compared from raw beef liver) are calculated based on the dry matter basis to take into account the amount of water in the product.

The placebo powder was also analysed in terms of its nutritional profiles (i.e., Vitamin A, proximates, and micronutrients), other trace elements and functional properties (i.e., colour, texture and flavour). It is the intention that the placebo powder should not contain or had only very little traces of Vitamin A, iron and zinc with similar functional properties to the dried beef liver powder and/or beef meat powder. The results in Table 8 confirmed that the placebo powder mainly contains carbohydrate (~97%) with very little amounts of Vitamin A, iron and zinc. The colour of the placebo powder was observed to be similar to dried beef liver powder (Fig. 14). It has a more free-flowing characteristic compared to the beef liver and meat powders due to the fact that it contains less fat. Moreover, the texture of the placebo powder is somewhat consistent to the beef liver powder and has a meaty flavour similar to the flavour of the beef meat powder.



(a)



(b)



(c)

Fig. 14. Photos of final dried powders (a) beef liver, (b) beef meat, and (c) placebo.

The manufacturing process for all dried powders was carried out in accordance with a HACCP (hazard analysis critical control point) food safety plan. This is in line with CSIRO's human health and medical research ethics with the approval from the Food Risk Assessment Team (FRAT) at CSIRO Food and Nutrition Flagship to ensure their fitness for human consumption. The process was carried out with good manufacturing practices (GMP) following the standard hygiene protocols for cleaning and operation of the processing equipment and accessories (Appendix 6). In addition, the powders were produced under food grade conditions (e.g., constant monitoring of product temperatures throughout the preparation and processing chain). The results of the microbiological tests for all dried

powders show that pathogens of potential concern were not detected and also the results for *Enterobacteriaceae*, *Salmonella*, Standard Plate Count, *Bacillus cereus*, *E.Coli* and so on were satisfactory (Table 10). These confirmed the compliance of the handling and processing conditions necessarily required for food safety and production of products that are fit for human consumption.

Table 10. Microbiological quality of the final dried powders.

Microbiological tests	Dried products			
	Beef liver powder	Beef meat powder	Beef meat & liver blend powder	Placebo powder
Salmonella	nd/25 g	nd/25 g	nd/25 g	nd/25 g
Standard Plate Count	<300 cfu/g	<1200 cfu/g	<300 cfu/g	<40 cfu/g
Enterobacteriaceae	<10 cfu/g	<10 cfu/g	<10 cfu/g	<10 cfu/g
B. cereus	<100 cfu/g	<100 cfu/g	<100 cfu/g	<100 cfu/g
Coagulase Positive Staph	<100 cfu/g	<100 cfu/g	<100 cfu/g	<100 cfu/g
E.Coli	<3.0 MPN/g	<3.0 MPN/g	<3.0 MPN/g	<3.0 MPN/g
Coliforms	<3.0 MPN/g	<3.0 MPN/g	3.6 MPN/g	<3.0 MPN/g
Staphylococcus aureus	<10 cfu/g	<10 cfu/g	<10 cfu/g	<10 cfu/g

Note: Blend ratio of liver powder to meat powder is 20:80. nd = not detected

Export Requirements

The dried powders are intended for nutritional trials in Indonesia. These products were therefore manufactured to comply with the regulatory requirements for export to this country. As a requirement for the nutritional study, these powders were finally packed into sachets. The details of the products and the corresponding sachet labelling and coding are presented in Table 11. The filling/sealing of the products into sachets was carried out by a contract food manufacturer based in Ballina, NSW (Australian Vitamin & Sports Nutrition Pty Ltd). The company's food manufacturing plant is operated under certified HACCP food safety program and Halal compliance, with staff fully trained in food safety and good manufacturing practices (GMP) (Appendix 7). Fig. 15 shows the final products in sachets with the details of the labelling and product coding.

Table 11. Details of sachet filling/sealing, product coding and labelling.

Parameters	Products			
	Beef liver powder	Beef meat powder	Beef meat & liver blend powder	Placebo powder
Product code*	1	2	3	4
Sticker label	Product 1	Product 2	Product 3	Product 4
Amount of powder in sachet (g)	15	20	20	15
Number of sachets produced	550	450	475	550

Note: * The product code for each product is directly printed in sachets. Blend ratio of liver powder to meat powder is 20:80.

A test shipment via FedEx of a small quantity of two dried products (i.e., 1 kg of beef liver powder and 1 kg of beef meat powder) from Australia to Indonesia was carried out to acquire a “pass bill” and to initially determine the export documentations required for the shipment of these products. The trial shipment was successful with the details of the required documentations formed the basis in the preparation of the documents required for the shipment of the final products (Appendix 8). Subsequently, an initial batch of these products was also successfully shipped to Indonesia, and the exportation of the remaining products is currently underway.



Fig. 15. Photo of the product in sachet with the labelling.

5 Conclusions and Recommendations

This project was commissioned for the manufacture of various dried powders (i.e., beef liver, beef meat and placebo) to the required quantity and specifications for nutritional trials in Indonesia. The main focus of the work was to develop a manufacturing process for the production of powdered dried beef liver that contains Vitamin A at the recommended level safe for intake (i.e., <6,000 µg per 100 g dried powder). The powder quality with respect to the nutritional, functional and microbiological specifications, and fitness for human consumption and suitability for export to Indonesia was determined.

A pilot scale manufacturing process was successfully developed and tested based from the processing methods and conditions established in the laboratory study, together with a series of pilot scale trials. The manufacturing process involved a series of food processing steps, consisting of major unit operations such as cooking in hot water, double step hot oil treatment, and freeze drying. The results from both laboratory and pilot trials show that up to 89% reduction in Vitamin A can be achieved through the process of dicing the raw fresh beef

liver into cubes (~10-15 mm), subsequently cooking at 80°C for 15-25 minutes and mincing followed by a double step hot oil treatment at 60°C for 2 hours in each step. This produced dried beef liver powder that contains the required level of Vitamin A. However, there were instances where the Vitamin A in the dried liver powder still exceeded the required level due to cases of extremely high levels of Vitamin A in raw fresh beef liver. In these cases, blending with dried meat powder was an ideal option to achieve the required level of Vitamin A as the dried meat powder was found to contain very low amounts of Vitamin A.

The findings from this work revealed a significant impact of the major processing steps in reducing the Vitamin A level. The cooking step was observed to contribute in the overall reduction of Vitamin A (7% reduction) and imparted a significant weight loss of the product (about 32%), in addition to its important role as a thermal treatment of the product for microbiological safety. The double hot oil treatment was found to be the main processing step to contribute to a major reduction (78%) of Vitamin A level in powdered beef liver and caused additional weight loss of 5%. The drying process contributed to the main reduction in weight (i.e., mainly moisture loss) of the product in the manufacturing process and resulted in a 10% reduction in Vitamin A.

The qualities of the dried powders produced at pilot scale were assessed in terms of their nutritional, functional and microbiological specifications. Both beef liver and meat powders were found to be a good source of protein and micronutrients (i.e., iron and zinc). The iron content of the beef liver powder was about 3 times higher than the beef meat powder. However, both micronutrients were found to be significantly affected by the manufacturing process. In addition, the placebo powder was also analysed in terms of its nutritional and functional properties and was found to contain very little traces of Vitamin A, iron and zinc (as intended) with similar functional properties to the dried beef liver powder and/or beef meat powder.

The pilot manufacturing process for the production of all dried powders was carried out in accordance with a HACCP food safety plan and good manufacturing practices to ensure their fitness for human consumption. The results of the microbiological tests of these dried powders confirmed the compliance of the manufacturing process with food safety requirements. These products were also manufactured to comply with the regulatory requirements for export to Indonesia. A trial shipment of these products to Indonesia, designed to acquire a “pass bill” and to determine the required export documentations, was successful, confirming their suitability for export to this country. Subsequently, a batch of the final products was also successfully shipped to Indonesia. The exportation of the remaining products is currently underway.

Overall, this work has generated new knowledge and invaluable insights in the manufacturing process of dried beef liver powder with the required level of Vitamin A. However, there are further issues that may be worthwhile investigating to fully strengthen adoption of the process at industrial operations. It should be noted that this work was limited to the development and application of a specific drying method (i.e., freeze drying), as the focus was mainly to produce and deliver a powdered dried beef liver, that is fit for human consumption and suitable for export. The high costs associated with the freeze drying method preclude the industrial scale utilisation of this technology.

It is therefore recommended that further R&D work is undertaken to establish fundamental understanding and basis to build upon the development of a cost-effective and scalable manufacturing process at industrial scale operations.

- Develop and optimise a new drying concept (e.g., application of ultrasonics) for a cost-effective and scalable process as an alternative to freeze drying to produce dried beef liver powder with reduced Vitamin A levels whilst minimising the impact of the process on micronutrients (i.e., iron and zinc) degradation with better control of the functional attributes (e.g., flowability, solubility) of the dried powder.
- Investigate the effects of pre/post drying processes in terms of process performance (i.e., cost-effectiveness, scalability and effectiveness in reducing Vitamin A levels) and their impact on micronutrients degradation, functional and microbial attributes of the dried product.
- Evaluate the possibility of masking the undesirable flavours of the dried product using microencapsulation and/or alternative masking technologies through improved understanding of the flavour profile as affected by processing using advanced techniques (e.g., GC-MS).
- Assess the potential of capturing Vitamin A in the extraction process as a further co-product (i.e., natural source of Vitamin A) and utilisation of other offals (e.g., heart, kidney, etc) for blending with the beef liver powder to alternatively achieve the required level of Vitamin A in a cost-effective manner.
- Study the stability of Vitamin A and micronutrients at different storage conditions in dried liver powder and in various food product formats fortified with dried liver powder, and develop strategies to minimise the degradation process during storage.

The findings from the future work would not only assist the red meat processing sector in enhancing their sustainability and profitability (i.e., through capturing more value from under-utilised co-products), but would also provide a readily accessible and affordable source of complimentary nutrient-rich food to address the global nutritional needs of the at-risk children populations (especially in developing countries). It is also expected that the manufacturing process could easily be extended to other meats and meat products.

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7 Appendix

Appendix 1. Halal certificates for beef liver and beef meat.

119, Aug. 2015 2:23 Magala: No. 249 P. 1/1



ISLAMIC CO-ORDINATING COUNCIL OF VICTORIA P/L
I.C.C.V



شهادة حلال
HALAL CERTIFICATE



Mutton / Lamb / Calves / Goats

The Islamic Co-ordinating Council of Victoria
hereby certify that **Wagstaff Cranbourne Pty Ltd. Est. No. 46**
is listed on the "Halal Approved Abattoir List". Therefore the
animals slaughtered by Muslim slaughtermen authorized by our organisation
using a knife and according to Islamic rites. The animals so slaughtered are Halal
and therefore suitable for consumption by Muslims in any part of the world.

An official Halal certificate signed and stamped by AQIS and I.C.C.V. for
every shipment is a Must.

Any consignment of meat without Halal Transfer Certificate must be
considered as Non Halal.

This certificate is valid until the **31st December 2015**
Authorized by: **Hafik Koya**
Inspection Manager

CERTIFICATE NO. : 13032015 / WC 15

ICCV reserves the right to inspect and audit this establishment at any time
without prior notice and reserves the right to cancel the accreditation should
there be any breach of ICCV Halal guidelines. This certificate remains the
property of ICCV and must be returned to ICCV upon request. A photocopy
of this certificate is not valid. This certificate is issued to Wagstaff Cranbourne
and is not to be photocopied or displayed by anyone else.

ABN: 30 542 393 936
TRUSTEE FOR THE HALAL BOARD OF AUSTRALIA TRUST
166 Lygon Street, East Brunswick, VIC 3087 Australia • PO Box 108, East Brunswick, VIC 3057
P: +61 3 9380 5457 F: +61 3 9380 6148 W: www.iccv.com.au E: iccv@bigpond.com

17/00/2015 08:42:00 Hongbo

(Fax) 03 52744766

P.001/001



ISLAMIC CO-ORDINATING COUNCIL OF VICTORIA P/L
I.C.C.V



شهادة حلال
HALAL CERTIFICATE



*** Beef / Mutton / Calves / Lamb ***

The Islamic Co-ordinating Council of Victoria

hereby certify that M. C. Herd Pty Ltd, Est. No. 13 is listed on the
"Halal Approved Abattoir List". Therefore the animals
slaughtered by Muslim slaughtermen authorized by our organisation using
a knife and according to Islamic rites. The animals so slaughtered are Halal
and therefore suitable for consumption by Muslims in any part of the world.

An official Halal certificate signed and stamped by AQIS and I.C.C.V.
for every shipment is a Must.

Any consignment of meat without Halal Transfer Certificate must be
considered as Non Halal.

This certificate is valid until the **31st December 2015**

Authorized by: Ehsan ul Haque
Accounts Manager & Halal Certifier

CERTIFICATE NO. : 2212014/ MCH 14

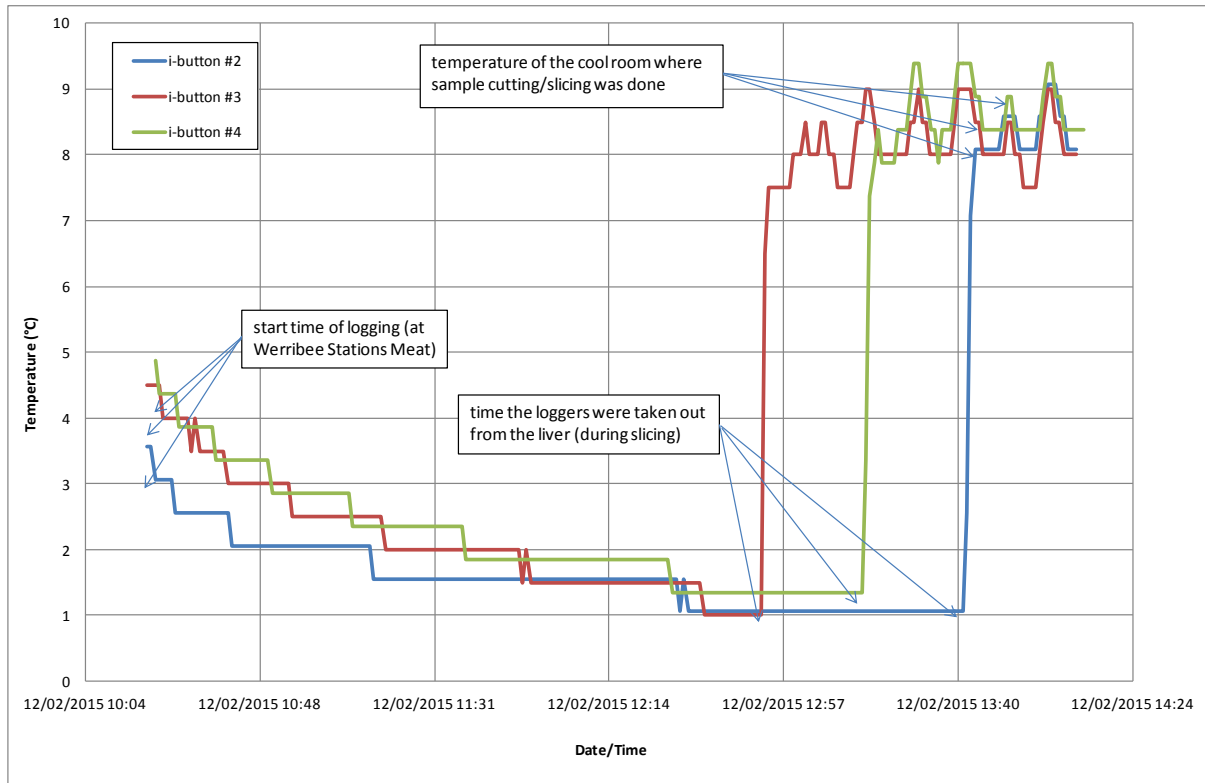
ICCV reserves the right to inspect and audit this establishment at any time without prior
notice and reserves the right to cancel the accreditation should there be any breach of
ICCV Halal guidelines. This certificate remains the property of ICCV and must be
returned to ICCV upon request. A photocopy of this certificate is not valid. This
certificate is issued to M. C. Herd and is not to be photocopied or displayed by anyone
else. **This certificate is not an export document.**

ABN: 50 542 393 936

TRUSTEE FOR THE HALAL BOARD OF AUSTRALIA TRUST

155 Lygon Street, East Brunswick, VIC 3057 Australia • PO Box 108, East Brunswick, VIC 3057
Ph +61 3 9380 5467 P: +61 3 9380 8143 W: www.iccv.com.au E: iccv@bigpond.com

Appendix 2. Example of the logged centre temperatures of the fresh beef liver during refrigerated transport.



Appendix 3. Product specification of the beef flavouring.



Product Specification

Interim

Product Code	XS4328
Product Name	BEEF FLAVOUR
GMO Status:	No GM Ingredients.
Allergens:	No Allergens.
Manufacturer:	Sensient Flavors (Wales) Ltd

Quality Parameters

Appearance:	Clear yellow liquid.
Taste & Odour:	Typical of beef. Free from off or objectionable flavours.
Regulatory Status:	Product complies with the Australia New Zealand Food Standards Code.
Kosher:	Compliant.
Halal:	Compliant.
Ingredients:	Flavouring.

Physical

Refractive Index:	1.456 - 1.476 at 20°C.
Specific Gravity:	0.897 - 0.917 at 20°C.
Flash Point:	>80°C (Closed Cup).

Date Printed: 05/03/15		Page 1 of 2 XS4328	
Date Issued:	Version:	Superseded:	Approved by: Technical Manager Quality Assurance Manager
05/03/15	1.0		

Technical information and suggestions for use, including any formulations and procedures are believed to be correct. However this does not constitute a guarantee of the accuracy of the information contained herein and confirming test in your own plant or laboratory is recommended. Whilst materials supplied by our firm may be legal in the country of origin, we do not warrant or guarantee their legality in other countries and highly recommend user confirmation.

Appendix 4. Proximates, Vitamin A, micronutrients and heavy metals analyses (NMI).



Australian Government

National Measurement Institute

REPORT OF ANALYSIS

Page: 1 of 2

Report No. RN1066518

Client :	CSIRO - FOOD & NUTRITIONAL SCIENCES	Job No. :	CSIRO54/150424
	671 SNEYDES ROAD	Quote No. :	QT-02039
	WERRIBEE VIC 3030	Order No. :	
		Date Sampled :	
Attention :	HENRY SABAREZ	Date Received :	24-APR-2015
Project Name :		Sampled By :	CLIENT
Your Client Services Manager :	Tim Stobaus	Phone :	(03) 9644 4849

Lab Reg No.	Sample Ref	Sample Description
V15/009789/1	BLA(1)	Beef liver powder-Apr batch(R1)
V15/009790/1	BLA(2)	Beef liver powder-Apr batch(R2)
V15/009791/1	BLF(1)	Beef liver powder-Feb batch(R1)
V15/009792/1	BLF(2)	Beef liver powder-Feb batch(R2)

Lab Reg No.		V15/009789/1	V15/009790/1	V15/009791/1	V15/009792/1	
Sample Reference	Units	BLA(1)	BLA(2)	BLF(1)	BLF(2)	Method
Proximates						
Fat (Mojonnier extraction)	g/100g	27.6	29.3	21.4	21.6	VL302
Protein (N x 6.25)	g/100g	48.8	55.0	59.4	65.3	VL299
Ash	g/100g	1.3	2.1	4.7	2.3	VL286
Carbohydrates	g/100g	3	4	13	9	VL412

Paul Adorno, Section Manager
Food Composition - Vic

11-MAY-2015

REPORT OF ANALYSIS

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Report No. RN1066518

Client	: CSIRO - FOOD & NUTRITIONAL SCIENCES 671 SNEYDES ROAD WERRIBEE VIC 3030	Job No.	: CSIRO54/150424
		Quote No.	: QT-02039
		Order No.	:
		Date Sampled	:
Attention	: HENRY SABAREZ	Date Received	: 24-APR-2015
Project Name	:	Sampled By	: CLIENT
Your Client Services Manager	: Tim Stobaus	Phone	: (03) 9644 4849

Lab Reg No.	Sample Ref	Sample Description
V15/009793/1	MLB(1)	Mince lean beef powder(R1)
V15/009794/1	MLB(2)	Mince lean beef powder(R2)
V15/009795/1	PL(1)	Placebo(R1)
V15/009796/1	PL(2)	Placebo(R2)

Lab Reg No.		V15/009793/1	V15/009794/1	V15/009795/1	V15/009796/1	
Sample Reference		MLB(1)	MLB(2)	PL(1)	PL(2)	
	Units					Method
Proximates						
Fat (Mojonnier extraction)	g/100g	22.3	22.4	<0.2	<0.2	VL302
Protein (N x 6.25)	g/100g	72.6	59.2	0.4	0.2	VL299
Ash	g/100g	1.1	2.1	<0.1	<0.1	VL286
Carbohydrates	g/100g	3	15	97	97	VL412



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11-MAY-2015

Results relate only to the sample(s) tested.
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National Measurement Institute

REPORT OF ANALYSIS

Page: 1 of 4
Report No. RN1066515

Client	: CSIRO - FOOD & NUTRITIONAL SCIENCES 671 SNEYDES ROAD WERRIBEE VIC 3030	Job No.	: CSIRO54/150424
		Quote No.	: QT-02039
		Order No.	:
		Date Sampled	:
		Date Received	: 24-APR-2015
Attention	: HENRY SABAREZ	Sampled By	: CLIENT
Project Name	:		
Your Client Services Manager	: Tim Stobaus	Phone	: (03) 9644 4849

Lab Reg No.	Sample Ref	Sample Description
V15/009789	BLA(1)	Beef liver powder-Apr batch(R1)
V15/009790	BLA(2)	Beef liver powder-Apr batch(R2)
V15/009791	BLF(1)	Beef liver powder-Feb batch(R1)
V15/009792	BLF(2)	Beef liver powder-Feb batch(R2)

Lab Reg No.		V15/009789	V15/009790	V15/009791	V15/009792	
Sample Reference		BLA(1)	BLA(2)	BLF(1)	BLF(2)	Method
	Units					
Trace Elements						
Iron	mg/kg	120	140	160	160	VL247
Zinc	mg/kg	110	120	150	150	VL247


Paul Adorno, Section Manager
Inorganics - Vic

11-MAY-2015

Lab Reg No.		V15/009789	V15/009790	V15/009791	V15/009792	
Sample Reference		BLA(1)	BLA(2)	BLF(1)	BLF(2)	Method
	Units					
Proximates						
Moisture	g/100g	19.7	10.1	1.4	1.6	VL298
Vitamins						
Retinol (Vitamin A)	ug/100g	44	4700	4300	4200	VL287


Paul Adorno, Section Manager
Food Composition - Vic


Norbert Strobel, Analyst
Food Composition - Vic

11-MAY-2015

REPORT OF ANALYSIS

Page: 2 of 4

Report No. RN1066515

Client	: CSIRO - FOOD & NUTRITIONAL SCIENCES	Job No.	: CSIRO54/150424
	671 SNEYDES ROAD	Quote No.	: QT-02039
	WERRIBEE VIC 3030	Order No.	:
		Date Sampled	:
Attention	: HENRY SABAREZ	Date Received	: 24-APR-2015
Project Name	:	Sampled By	: CLIENT
Your Client Services Manager	: Tim Stobaus	Phone	: (03) 9644 4849

Lab Reg No.	Sample Ref	Sample Description
V15/009793	MLB(1)	Mince lean beef powder(R1)
V15/009794	MLB(2)	Mince lean beef powder(R2)
V15/009795	PL(1)	Placebo(R1)
V15/009796	PL(2)	Placebo(R2)

Lab Reg No.		V15/009793	V15/009794	V15/009795	V15/009796	
Sample Reference		MLB(1)	MLB(2)	PL(1)	PL(2)	Method
	Units					
Trace Elements						
Iron	mg/kg	46	45	<2	<2	VL247
Zinc	mg/kg	180	180	0.30	0.23	VL247


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11-MAY-2015

Lab Reg No.		V15/009793	V15/009794	V15/009795	V15/009796	
Sample Reference		MLB(1)	MLB(2)	PL(1)	PL(2)	Method
	Units					
Proximates						
Moisture	g/100g	1.3	1.5	2.6	2.7	VL298
Vitamins						
Retinol (Vitamin A)	ug/100g	23	21	<5	<5	VL287


 Paul Adorno, Section Manager
 Food Composition - Vic


 Norbert Strobel, Analyst
 Food Composition - Vic

11-MAY-2015

REPORT OF ANALYSIS

Page: 3 of 4
Report No. RN1066515

Client	: CSIRO - FOOD & NUTRITIONAL SCIENCES 671 SNEYDES ROAD WERRIBEE VIC 3030	Job No.	: CSIRO54/150424
		Quote No.	: QT-02039
		Order No.	:
		Date Sampled	:
		Date Received	: 24-APR-2015
Attention	: HENRY SABAREZ	Sampled By	: CLIENT
Project Name	:		
Your Client Services Manager	: Tim Stobaus	Phone	: (03) 9644 4849

Lab Reg No.	Sample Ref	Sample Description
V15/009797	BLF-Mix(1)	Beef liver powder- mix Feb batch(R1)
V15/009798	BLF-Mix(2)	Beef liver powder- mix Feb batch(R2)
V15/009799	R1	Raw Beef liver (R1)
V15/009800	R2	Raw Beef liver (R2)

Lab Reg No.		V15/009797	V15/009798	V15/009799	V15/009800	
Sample Reference		BLF-Mix(1)	BLF-Mix(2)	R1	R2	Method
	Units					
Trace Elements						
Iron	mg/kg	160	160	83	83	VL247
Zinc	mg/kg	160	160	83	83	VL247



Paul Adorno, Section Manager
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11-MAY-2015

Lab Reg No.		V15/009797	V15/009798	V15/009799	V15/009800	
Sample Reference		BLF-Mix(1)	BLF-Mix(2)	R1	R2	Method
	Units					
Proximates						
Moisture	g/100g	1.4	1.3	71.2	70.7	VL298
Vitamins						
Retinol (Vitamin A)	ug/100g	4300	4500	12000	18000	VL287



Paul Adorno, Section Manager
Food Composition - Vic



Norbert Strobel, Analyst
Food Composition - Vic

11-MAY-2015



Australian Government
Department of Industry and Science

National
Measurement
Institute

REPORT OF ANALYSIS

Page: 1 of 1
Report No. RN1078782

Client	: CSIRO - FOOD & NUTRITIONAL SCIENCES 671 SNEYDES ROAD WERRIBEE VIC 3030	Job No.	: CSIRO54/150804
		Quote No.	: QT-02039
		Order No.	:
		Date Sampled	:
		Date Received	: 4-AUG-2015
Attention	: HENRY SABAREZ	Sampled By	: CLIENT
Project Name	:		
Your Client Services Manager	: Tim Stobaus	Phone	: (03) 9644 4849

Lab Reg No.	Sample Ref	Sample Description
V15/022129	Product 1	Beef liver powder(x2 Sachets)
V15/022130	Product 2	Beef meat powder(x2 Sachets)
V15/022131	Product 3	Beef liver /beef meat blend powder(x2 Sachets)
V15/022132	Product 4	Placebo powder(x2 Sachets)

Lab Reg No.		V15/022129	V15/022130	V15/022131	V15/022132	
Sample Reference		Product 1	Product 2	Product 3	Product 4	
	Units					Method
Trace Elements						
Antimony	mg/kg	<0.01	<0.01	<0.01	<0.01	VL247
Arsenic	mg/kg	0.020	0.032	0.028	<0.01	VL247
Cadmium	mg/kg	0.087	<0.01	0.018	<0.01	VL247
Copper	mg/kg	61	2.6	15	0.089	VL247
Lead	mg/kg	0.070	0.013	0.026	0.017	VL247
Mercury	mg/kg	<0.01	<0.01	<0.01	<0.01	VL247
Selenium	mg/kg	0.48	0.20	0.28	<0.01	VL247
Tin	mg/kg	0.27	0.024	0.048	<0.01	VL247
Zinc	mg/kg	130	180	170	1.0	VL247


Paul Adorno, Section Manager
Inorganics - Vic

14-AUG-2015

Appendix 5. Microbiological tests of the dried powders (DTS Pty Ltd).

(a) Beef liver powder



Dairy Technical Services Ltd
ABN 30 004 319 171
Corporate Office
3/63-71 Boundary Road
North Melbourne VIC
Australia 3061
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Tel: +61 (3) 8371 7600
Fax: +61 (3) 9372 2013

Postal Address
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Microbiology
Laboratory Victoria
3/352 Macaulay Road
Kensington VIC 3031

Chemistry Laboratory
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North Melbourne
VIC 3051

Microbiology and
FACTA Allergies
Laboratory Queensland
Unit 1-3/148 Tennyson
Memorial Avenue
Tennyson QLD 4103
Tel: +61 (7) 3426 9750
Fax: +61 (7) 3392 8495

LABORATORY REPORT

on

MEAT AND BONE MEAL

Date: 27/04/2015

Our Ref: DTS15030501

Report No: 1512967

Final

FOR: CSIRO CFNS WERRIBEE

671 Sneydes Rd
Werribee VIC

3030

Sieh Ng

Date received: 23/04/2015

Origin:

Code/Ref: CSIRO BEEF LIVER POWDER 01/04
/15

Order Number:

No of samples: 6

Package Type:

Temperature on receipt: Ambient

TEST	RESULTS	METHOD N
23APR15/7321189 Client ID: 13. BEEF LIVER POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321190 Client ID: 14. BEEF LIVER POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321192 Client ID: 15. BEEF LIVER POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321193 Client ID: 16. BEEF LIVER POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321195 Client ID: 17. BEEF LIVER POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321196 Client ID: 18. BEEF LIVER POWDER 01/04/15		
Standard Plate Count	240 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06

Kathleen Cleary
Technical Specialist



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LABORATORY REPORT
on
MEAT AND BONE MEAL

Date: 22/05/2015
Our Ref: DTS15036653
Report No: 1532902
Final

FOR: CSIRO CFNS WERRIBEE

671 Sneydes Rd
Werribee VIC

3030

Sieh Ng

Date received: 14/05/2015

Origin:

Code/Ref: CSIRO LIVER POWDER 01.05.15

Order Number:

No of samples: 5

Package Type:

Temperature on receipt: Ambient

TEST	RESULTS	METHOD N
14MAY15/7392401		
Client ID: 1. LIVER POWDER 01.04.15		
Standard Plate Count	150 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
Yeasts	<100 cfu/g	YMFD 01 05.05
Moulds	<100 cfu/g	YMFD 01 05.05
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06
14MAY15/7392413		
Client ID: 2. LIVER POWDER 01.04.15		
Standard Plate Count	120 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
Yeasts	<100 cfu/g	YMFD 01 05.05
Moulds	<100 cfu/g	YMFD 01 05.05
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06
14MAY15/7392416		
Client ID: 3. LIVER POWDER 01.04.15		
Standard Plate Count	110 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
Yeasts	<100 cfu/g	YMFD 01 05.05
Moulds	<100 cfu/g	YMFD 01 05.05
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06



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LABORATORY REPORT
on
MEAT AND BONE MEAL

Date: 22/05/2015
Our Ref: DTS15036653
Report No: 1532902
Final

FOR: CSIRO CFNS WERRIBEE

671 Sneydes Rd
Werribee VIC 3030

Sieh Ng

Date received: 14/05/2015

Origin:

Code/Ref: CSIRO LIVER POWDER 01.05.15

Order Number:

No of samples: 5

Package Type:

Temperature on receipt: Ambient

TEST	RESULTS	METHOD N
14MAY15/7392417		
Client ID: 4. LIVER POWDER 01.04.15		
Standard Plate Count	140 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
Yeasts	<100 cfu/g	YMFD 01 05.05
Moulds	<100 cfu/g	YMFD 01 05.05
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06
14MAY15/7392418		
Client ID: 5. LIVER POWDER 01.04.15		
Standard Plate Count	60 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
Yeasts	<100 cfu/g	YMFD 01 05.05
Moulds	<100 cfu/g	YMFD 01 05.05
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06

Marygrace Navarro
Microbiologist



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LABORATORY REPORT
on
MEAT PRODUCTS

Date: 10/08/2015
Our Ref: DTS15058977
Report No: 1593030
Final

FOR: CSIRO CFNS WERRIBEE

671 Sneydes Rd
Werribee VIC 3030

Sieh Ng

Date received: 5/08/2015
Origin:
Code/Ref: SAMPLE 1-3 BEEF POWDERS
Order Number:
No of samples: 3
Package Type:
Temperature on receipt: Ambient

TEST	RESULTS	METHOD N
05AUG15/7642861		
Client ID: 1. BEEF LIVER POWDER 20/04/15		
Standard Plate Count	300 cfu/g	PCFD 04 10.05
Coliforms	< 3.0 MPN/g	CEFD 02.05 05
E.Coli	< 3.0 MPN/g	CEFD 02.05 05
B. cereus	<100 cfu/g	BCFD 04 08.08
Staphylococcus aureus	<10 cfu/g	STFD 08 07.13
Salmonella	Not Detected /25g	SMFD 02 05.05
05AUG15/7642862		
Client ID: 2. BEEF MEAT POWDER 14/05/15		
Standard Plate Count	1200 cfu/g	PCFD 04 10.05
Coliforms	< 3.0 MPN/g	CEFD 02.05 05
E.Coli	< 3.0 MPN/g	CEFD 02.05 05
B. cereus	<100 cfu/g	BCFD 04 08.08
Staphylococcus aureus	<10 cfu/g	STFD 08 07.13
Salmonella	Not Detected /25g	SMFD 02 05.05
05AUG15/7642863		
Client ID: 3. BEEF LIVER & MEAT MIXTURE POWDER 15/05/15		
Standard Plate Count	300 cfu/g	PCFD 04 10.05
Coliforms	3.6 MPN/g	CEFD 02.05 05
E.Coli	< 3.0 MPN/g	CEFD 02.05 05
B. cereus	<100 cfu/g	BCFD 04 08.08
Staphylococcus aureus	<10 cfu/g	STFD 08 07.13
Salmonella	Not Detected /25g	SMFD 02 05.05

Kathleen Cleary
Technical Specialist

(b) Beef meat powder



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LABORATORY REPORT
on
MEAT AND BONE MEAL

Date: 27/04/2015
Our Ref: DTS15030493
Report No: 1512965
Final

FOR: CSIRO CFNS WERRIBEE

671 Sneydes Rd
Werribee VIC 3030

Sieh Ng

Date received: 23/04/2015

Order Number:

Origin:

No of samples: 6

Code/Ref: CSIRO BEEF MINCE POWDER 01/
04/15

Package Type:

Temperature on receipt: Ambient

TEST	RESULTS	METHOD N
23APR15/7321091 Client ID: 7. BEEF MINCE POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321093 Client ID: 8. BEEF MINCE POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321094 Client ID: 9. BEEF MINCE POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321095 Client ID: 10. BEEF MINCE POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321097 Client ID: 11. BEEF MINCE POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321098 Client ID: 12. BEEF MINCE POWDER 01/04/15		
Standard Plate Count	20 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06

Kathleen Cleary
Technical Specialist

(c) Placebo powder



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LABORATORY REPORT

on

MALTODEXTRIN

Date: 26/04/2015
Our Ref: DTS15030485
Report No: 1512533
Final

FOR: CSIRO CFNS WERRIBEE

671 Sneydes Rd
Werribee VIC 3030

Sieh Ng

Date received: 23/04/2015

Origin:

Code/Ref: CSIRO MALTODEXTRIN POWDER
01/04/15

Order Number:

No of samples: 6

Package Type:

Temperature on receipt: Ambient

TEST	RESULTS	METHOD N
23APR15/7321052		
Client ID: 1. MALTODEXTRIN POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321053		
Client ID: 2. MALTODEXTRIN POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321054		
Client ID: 3. MALTODEXTRIN POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321055		
Client ID: 4. MALTODEXTRIN POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321056		
Client ID: 5. MALTODEXTRIN POWDER 01/04/15		
Salmonella	Not Detected /25g	SMFD 02 05.05
23APR15/7321065		
Client ID: 6. MALTODEXTRIN POWDER 01/04/15		
Standard Plate Count	40 cfu/g	PCFD 04 10.05
Enterobacteriaceae	<10 cfu/g	ENFD 12 12.08
B. cereus	<100 cfu/g	BCFD 04 08.08
Coagulase Positive Staph	<100 cfu/g	STFD 03 09.06

Abraham Gut
Microbiologist



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LABORATORY REPORT
on
MALTODEXTRIN

Date: 09/08/2015
Our Ref: DTS15058981
Report No: 1591922
Final

FOR: CSIRO CFNS WERRIBEE

671 Sneydes Rd
Werribee VIC 3030

Sieh Ng

Date received: 5/08/2015	Order Number:
Origin:	No of samples: 3
Code/Ref: SAMPLE 4 MALTODEXTRIN POWDER 29/03/15	Package Type:
Temperature on receipt: Ambient	

TEST	RESULTS	METHOD N
05AUG15/7642897		
Client ID: SAMPLE 4 MALTODEXTRIN POWDER 29/03/15		
Standard Plate Count	20 cfu/g	PCFD 04 10.05
Coliforms	< 3.0 MPN/g	CEFD 02.05 05
E.Coli	< 3.0 MPN/g	CEFD 02.05 05
B. cereus	<100 cfu/g	BCFD 04 08.08
Staphylococcus aureus	<10 cfu/g	STFD 08 07.13
Salmonella	Not Detected /25g	SMFD 02 05.05

Abraham Gut
Microbiologist

Appendix 6. Sample documents of the (a) CSIRO's FRAT approval, and (b) an example of record sheet for biotrace surface swabbing to monitor hygiene cleaning of the equipment and accessories.

CSIRO FNF Risk Assessment Process for Foods for Human Consumption

Risk Assessment for foods for human consumption

	Project number, name and Project Leader: PNO/Name: R-05157-13 / Powdered desiccated liver preparation PL: Henry Sabarez	PL Initial & status/done (Y/N/NA)
Step 1:	PL to notify Food Risk Assessment Team (FRAT) that project involves human consumption. Include project timing (see page 2)	Y
Step 2:	a. Page 4 – PL to draw flow diagram of proposed process noting location, timeframes and intended process temperatures for stages/steps. b. Page 5 – PL to fill in product characterisation form c. PL to forward to FRAT for risk level indication from FRAT	Y
Step 3:	FRAT to review and use the risk assessment decision tree – starting from Table A choose the table which best reflects the product you are producing and work through the Decision Tree recording the final path number on the product characterisation form. If a product falls outside any of the options of the decision tree the level of risk assessment will be determined by the FRAT.	Y
Step 4:	If a low level risk is indicated, flow diagram and product characterisation form submitted to the FRAT for checking and sign off. If a medium level risk is indicated, flow diagram and product characterisation form submitted to the FRAT for review before sign off and follow any actions required or recommendations they make. If a high level risk is indicated, a hazard analysis by the FRAT is required before sign off. Provide and submit documentation requested by the FRAT team. Working with the FRAT team, determine suitable actions to manage the risk to the consumer.	High level risk (Indicated path 4.3.1 Using decision tree v 12)
Step 5:	FRAT to return signed approval to proceed with required actions to PL	23 Feb 2015
Step 6:	PL to ensure that all local procedures are adhered to.	
Step 7:	PL to file all records and documents from the Risk Assessment with any hard copy or electronic project files.	
Step 8:	PL to send scanned copy of PL signed FRAT document to FRAT on project completion or at close of the approved human consumption phase.	

HIGH LEVEL RISK is indicated as summarised below and described in further detail on the following pages

Requires FRAT Hazard Analysis and appropriate actions.

1. Time/temperature control during product preparation to be logged on control sheets for each product.
2. **CORE** temperature of liver must reach >75°C.
3. Any change to product formulation, processing, shelf life etc will require a new risk assessment
4. All recommendation made **MUST** be adhered to and documented, prior to final authorisation

ACTIONS REQUIRED BY PROJECT TEAM ARE DETAILED OVER

Authorisation* by FRAT	Name	Signature	Date
Risk assessment reviewer 1	Sieh Ng	<i>Ng Sieh Yee</i>	11 Feb 2015
Risk assessment reviewed and approved by	Cathy Moir	<i>Cathy Moir</i>	23 Feb 2015

* This acknowledges that a food safety risk assessment has been completed based on the information provided by the project leader and the FRAT recommendations and actions required by the project leader are satisfactory to manage the risk to the consumer to a reasonably practicable level. The FRAT are not authorised to provide certificates of clearance for products.

RA-C03_2015 FRAT approval liver powder 20150223.docx

Page 1 of 5

Template updated by C. Moir 010714 CSIRO Food and Nutrition Flagship INTERNAL USE ONLY

(a)

Appendix 3

Daily Record Sheet

Operator: *Henry/Danny/Michael* Date: *01/04/15*
Site: *FFC Melaleuca 1* Shift: *8.55 am*

Insert Biotrace reading into appropriate column

SWAB No.	LOCATION (CCP)	PASS	ACCEPT LEVEL	CAUTION	REJECT LEVEL	FAIL	COMMENTS
0	CONTROL						
1	Iron						
2	vast 1						
3	temp probe	48					
4	bucket 1	81					
5	bucket 2	55					
6	bucket 3	15					
7	bucket 4	8					
8	spice bucket 1	8					
9	big pit	6					
10	spice bucket 2	11					
11	spice bucket 3	13					
12	Kettle 1	10					
13	Kettle 2	14					
14	chopped board	30					
15	spice (clay) 1	32					
16	spice (clay) 2	40					
17	oil probe	14					
18	temp probe 2	14					
19	food Kettle	7					
20	blade - mixer	7					
21	tray	10					
22	spice bucket 1	14					
23	mixer die	12					
24	mixer mixer	9					
25	food mixer	13					
26	mixer feed	15					
27	mixer outlet	81					

Checked By: *Sieh Ng* Signature: *Ng Sieh Yee* Date: *01/04/15*

Comments: The items that failed were re-cleaned and they passed the hygiene test on subsequent tests.

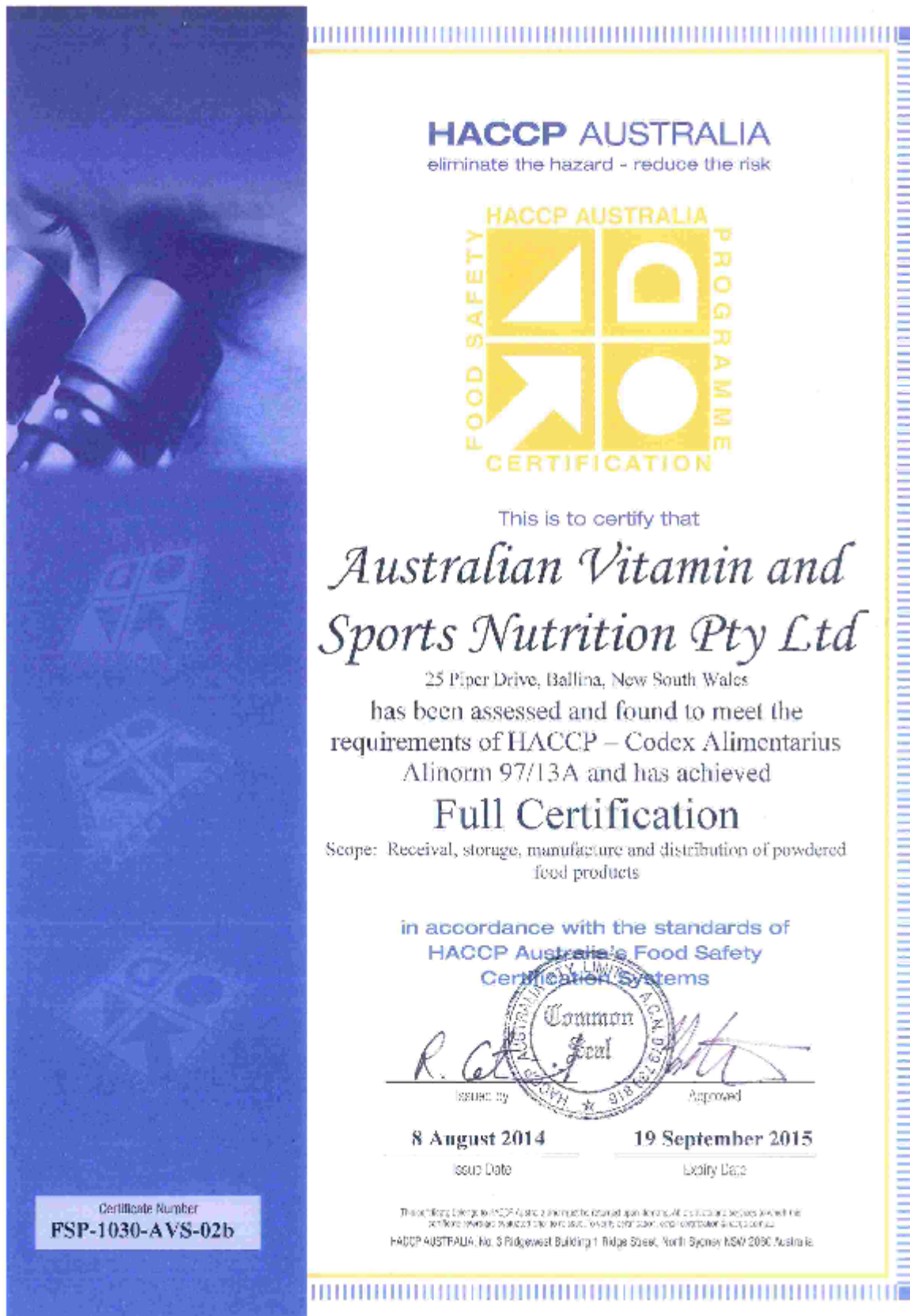
(28) plunger 13
(29) reclean vast 1 11

BIOTRACE
ENTERAL SYSTEMS
www.biotrace.com

Issue: 001 Issue Ref: APN003 (Formerly MLM 0475) (Copy UNCONTROLLED DOCUMENT 29/07/2003)

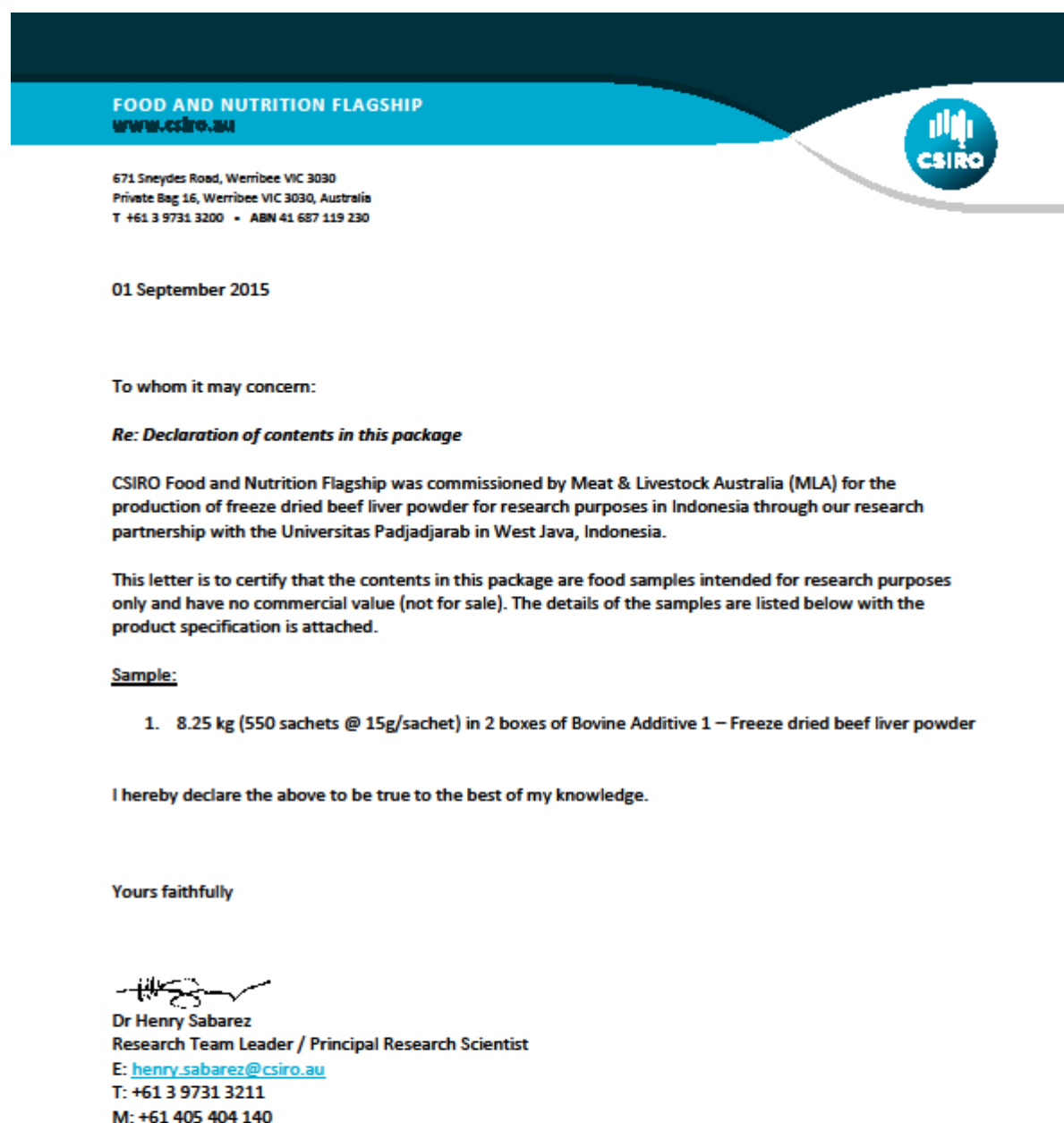
(b)

Appendix 7. HACCP certification of the Australian Vitamin & Sports (AVS) Nutrition Pty Ltd.



Appendix 8. Export documentations of the final products.

(a) Letters of declaration for the shipments



FOOD AND NUTRITION FLAGSHIP
www.csiro.au



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Private Bag 16, Werribee VIC 3030, Australia
T +61 3 9731 3200 • ABN 41 687 119 230

05 October 2015

To whom it may concern:

Re: Declaration of contents in this package

CSIRO Food and Nutrition Flagship was commissioned by Meat & Livestock Australia (MLA) for the production of freeze dried beef liver powder for research purposes in Indonesia through our research partnership with the Universitas Padjadjarab in West Java, Indonesia.

This letter is to certify that the contents in this package are food samples intended for research purposes only and have no commercial value (not for sale). The samples are listed below with the details of product specification for each sample are attached.

Samples:

1. 9.00 kg (450 sachets @ 20g/sachet) in 2 boxes of Bovine Additive 2 – Freeze dried beef liver powder
2. 9.50 kg (475 sachets @ 20g/sachet) in 2 boxes of Bovine Additive 3 - Freeze dried beef meat & liver blend powder
3. 8.25 kg (550 sachets @ 15g/sachet) in 2 boxes of Placebo – Freeze dried maltodextrin (with added beef-based flavouring & colouring) powder

I hereby declare the above to be true to the best of my knowledge.

Yours faithfully

A handwritten signature in black ink, appearing to read "Henry Sabarez".

Dr Henry Sabarez
Research Team Leader / Principal Research Scientist
E: henry.sabarez@csiro.au
T: +61 3 9731 3211
M: +61 405 404 140

(b) Commercial invoices for the shipments



COMMERCIAL INVOICE

AIRWAYBILL NO: ABN Number 4168119230		DATE OF EXPORTATION:	
EXPORTER/SHIPPER Company Name: CSIRO Food and Nutrition Flagship		INVOICE NO: REF NO: CONSIGNEE Dr Aly Diana Company Name: Universitas Padjadjaran	
Address: CSIRO Food and Nutrition Flagship 671 Sneydes road Wentree VIC 3030 Australia PH 64737313482 Fax 6473027313205		Address: Department of Medical Nutrition, Faculty of Medicine Jln Eljiman No. 38 Bandung 40151, West Java, Indonesia Tel: +62 81573100503	
COUNTRY OF EXPORT Australia		MANUFACTURER'S NAME (if not Shipper) Address: Shipper	
COUNTRY OF ULTIMATE DESTINATION Indonesia			

ITEMS	FULL DESCRIPTION OF GOODS	QTY	PART #	COUNTRY OF MFR	ARECC CODE	UNIT VALUE	TOTAL VALUE
1	Bovine Additive 1 - Freeze dried beef liver powder (250 sachets @ 15g/sachet) in 2 boxes	5.25kg		Australia			25 AUD

PLEASE TICK ONE: FCA CPT CIP

PLEASE STATE IF GOODS ARE DUTY DRAWBACK N

PLEASE STATE IF GOODS ARE HAZARDOUS N

REASON FOR EXPORT: Food supplement samples for research purposes (i.e. nutritional studies)

PERMIT NO: (If applicable) ENCRYPTION CODE: (If applicable)

**** Security Declarations: MUST BE SIGNED BELOW

I declare all the above information to be true and correct to the best of my knowledge and that the goods are of the origin specified above.

Also, "As a representative of the company named below, I confirm that we are the owner or originator of cargo we present for carriage, and confirm that the cargo is prepared and handled in a manner which will not compromise its security standing"

FOR & ON BEHALF OF:

COMPANY: CSIRO Food and Nutrition Flagship

NAME: (in print) Henry Sabarez

POSITION: Research Team Leader / Principal Research Scientist

SIGNATURE: 

DATE: 01/09/2015

GST: FREIGHT: INSURANCE: CURRENCY: GRND TOTAL: \$ 25.00 AUD



COMMERCIAL INVOICE

AIRWAYBILL NO: ASN Number 4168110230		DATE OF EXPORTATION:	
EXPORTER/SHIPPER Company Name: CSIRO Food and Nutrition Flagship Address: CSIRO Food and Nutrition Flagship 671 Sneydes road Werribee VIC 3030 Australia PH 643987313402 Fax 643987313205		CONSIGNEE Dr Aly Diana Company Name: Universitas Padjadjaran Address: Department of Medical Nutrition, Faculty of Medicine Jln Ejlaman No. 38 Bandung 40161, West Java, Indonesia Tel: +62 8121 4808681	
COUNTRY OF EXPORT Australia		MANUFACTURER'S NAME (if not Shipper) Address:	
COUNTRY OF ULTIMATE DESTINATION Indonesia		Shipper	

ITEMS	FULL DESCRIPTION OF GOODS	QTY	PART #	COUNTRY OF MFR	HS CODE	UNIT VALUE	TOTAL VALUE
1	Bovine Additive 2 - Freeze dried beef meat powder (400 sachets @ 20g/sachet) in 2 boxes	800g		Australia			10 AUD
2	Bovine Additive 3 - Freeze dried beef meat & liver blend powder (475 sachets @ 20g/sachet) in 2 boxes	950g		Australia			10 AUD
3	Placebo - Freeze dried maltodextrin (with added beef-based flavouring & colouring) powder (250 sachets @ 15g/sachet) in 2 boxes	3750g		Australia			10 AUD

PLEASE TICK ONE:

FCA	N	GST:
CPT	N	FREIGHT:
CIP		INSURANCE:
		CURRENCY:
		GRND TOTL:

PLEASE STATE IF GOODS ARE DUTY DRAWBACK

PLEASE STATE IF GOODS ARE HAZARDOUS

REASON FOR I Food supplement samples for research purposes only (i.e. nutritional studies) & not for sale

PERMIT NO: (if applicable)

ENCRYPTION CODE: (if applicable)

Security Declarations: MUST BE SIGNED BELOW

I declare all the above information to be true and correct to the best of my knowledge and that the goods are of the origin specified above.

Also, "As a representative of the company named below, I confirm that we are the owner or originator of cargo we present for carriage, and confirm that the cargo is prepared and handled in a manner which will not compromise its security standing"

FOR & ON BEHALF OF:

COMPANY: CSIRO Food and Nutrition Flagship

NAME: (in print) Henry Sabanez

POSITION: Research Team Leader / Principal Research Scientist

SIGNATURE:

DATE: 05/10/2015

(c) Product specification of the beef liver powder

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 Private Bag 16, Werribee VIC 3030, Australia
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Product Specification

Product: Bovine Additive 1
Date of Manufacture: 25 February 2015

Description: Beef liver powder is a freeze dried powder processed from Halal certified edible bovine livers. This product appears as a brownish powder.

Process: Frozen raw beef livers are sliced, cooked (80°C for 20 min), minced, heated and agitated in vegetable oil, rinsed with hot water, and frozen. The frozen beef liver is freeze dried, milled, sieved, and blended to meet the specifications.

Specifications:

<u>Proximates:</u>			
Moisture			1.5%
Fat			21.5%
Protein			62.3%
Ash			3.5%
Carbohydrate			11%
<u>Vitamin:</u>			
Vitamin A			4250 µg/100g
<u>Trace Elements:</u>			
Iron	160 mg/kg	Zinc	150 mg/kg
Antimony	<0.01 mg/kg	Arsenic	0.02 mg/kg
Cadmium	0.087 mg/kg	Copper	61 mg/kg
Lead	0.07 mg/kg	Mercury	<0.01 mg/kg
Selenium	0.48 mg/kg	Tin	0.27 mg/kg
<u>Bacteriological:</u>			
Salmonella			not detected /25g
Standard Plate Count			<300 cfu/g
Enterobacteriaceae			<10 cfu/g
B. cereus			<100 cfu/g
Coagulase Positive Staph			<100 cfu/g
E.Coli			<3.0 MPN/g
Coliforms			<3.0 MPN/g
Staphylococcus aureus			<10 cfu/g

Microbiological result*:
 The results of the microbiological analysis are provided above. These results apply to powder which was produced under food grade conditions as described here. The results show that pathogens of potential concern were not detected and results for Enterobacteriaceae and total counts were satisfactory; therefore indicate that the powder as tested is suitable for human consumption (*confirmed and approved by the Food Risk Assessment Team, CSIRO Food & Nutrition Flagship).

Comments:
 All ingredients used in the manufacture of these samples are food grade. The powder was prepared under a food safety plan with good manufacturing practices at CSIRO Food & Nutrition Flagship Werribee pilot plant facility.

Disclaimer:
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1 of 1

(d) Product specification of the beef meat powder

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671 Sneydes Road, Werribee VIC 3030
Private Bag 16, Werribee VIC 3030, Australia
T +61 3 9731 3200 • ABN 41 687 119 230

Product Specification

Product:	Bovine Additive 2																																																														
Date of Manufacture:	30 March 2015																																																														
Description:	Beef meat powder is a freeze dried powder processed from Halal certified edible minced beef meat. This product appears as a brownish powder.																																																														
Process:	Chilled raw minced beef meat are cooked (80°C for 15 min), rinsed with hot water, and frozen. The frozen beef is freeze dried, milled, sieved, and blended to meet the specifications.																																																														
Specifications:	<u>Proximates:</u> <table style="width: 100%;"> <tr><td>Moisture</td><td></td><td></td><td>1.4%</td></tr> <tr><td>Fat</td><td></td><td></td><td>22.4%</td></tr> <tr><td>Protein</td><td></td><td></td><td>65.9%</td></tr> <tr><td>Ash</td><td></td><td></td><td>1.6%</td></tr> <tr><td>Carbohydrate</td><td></td><td></td><td>9%</td></tr> </table> <u>Vitamin:</u> <table style="width: 100%;"> <tr><td>Vitamin A</td><td></td><td></td><td>22 µg/100g</td></tr> </table> <u>Trace Elements:</u> <table style="width: 100%;"> <tr> <td>Iron</td><td>45 mg/kg</td> <td>Zinc</td><td>180 mg/kg</td> </tr> <tr> <td>Antimony</td><td><0.01 mg/kg</td> <td>Arsenic</td><td>0.032 mg/kg</td> </tr> <tr> <td>Cadmium</td><td><0.01 mg/kg</td> <td>Copper</td><td>2.6 mg/kg</td> </tr> <tr> <td>Lead</td><td>0.013 mg/kg</td> <td>Mercury</td><td><0.01 mg/kg</td> </tr> <tr> <td>Selenium</td><td>0.2 mg/kg</td> <td>Tin</td><td>0.024 mg/kg</td> </tr> </table> <u>Bacteriological:</u> <table style="width: 100%;"> <tr><td>Salmonella</td><td>not detected /25g</td></tr> <tr><td>Standard Plate Count</td><td><1200 cfu/g</td></tr> <tr><td>Enterobacteriaceae</td><td><10 cfu/g</td></tr> <tr><td>B. cereus</td><td><100 cfu/g</td></tr> <tr><td>Coagulase Positive Staph</td><td><100 cfu/g</td></tr> <tr><td>E.Coli</td><td><3.0 MPN/g</td></tr> <tr><td>Coliforms</td><td><3.0 MPN/g</td></tr> <tr><td>Staphylococcus aureus</td><td><10 cfu/g</td></tr> </table>			Moisture			1.4%	Fat			22.4%	Protein			65.9%	Ash			1.6%	Carbohydrate			9%	Vitamin A			22 µg/100g	Iron	45 mg/kg	Zinc	180 mg/kg	Antimony	<0.01 mg/kg	Arsenic	0.032 mg/kg	Cadmium	<0.01 mg/kg	Copper	2.6 mg/kg	Lead	0.013 mg/kg	Mercury	<0.01 mg/kg	Selenium	0.2 mg/kg	Tin	0.024 mg/kg	Salmonella	not detected /25g	Standard Plate Count	<1200 cfu/g	Enterobacteriaceae	<10 cfu/g	B. cereus	<100 cfu/g	Coagulase Positive Staph	<100 cfu/g	E.Coli	<3.0 MPN/g	Coliforms	<3.0 MPN/g	Staphylococcus aureus	<10 cfu/g
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Microbiological result*:
The results of the microbiological analysis are provided above. These results apply to powder which was produced under food grade conditions as described here. The results show that pathogens of potential concern were not detected and results for Enterobacteriaceae and total counts were satisfactory; therefore indicate that the powder as tested is suitable for human consumption (*confirmed and approved by the Food Risk Assessment Team, CSIRO Food & Nutrition Flagship).

Comments:
All ingredients used in the manufacture of these samples are food grade. The powder was prepared under a food safety plan with good manufacturing practices at CSIRO Food & Nutrition Flagship Werribee pilot plant facility.

Disclaimer:
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(e) Product specification of the beef meat and liver blend powder

**Product Specification**

Product: Bovine Additive 3
Date of Manufacture: 30 March 2015

Description: A 20:80 blend of Bovine Additive 1 product (beef liver powder) and Bovine Additive 2 product (beef meat powder).

Process: Both products were processed separately as detailed in Bovine Additive 1 product (beef liver powder) and in Bovine Additive 2 product (beef meat powder).

Specifications:**Proximates:**

Moisture	2.6%
Fat	19.2%
Protein	71.2%
Ash	7.1%
Carbohydrate	<1.0%

Vitamin:

Vitamin A	175 µg/100g
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Trace Elements:

Iron	61.5 mg/kg	Zinc	155 mg/kg
Antimony	<0.01 mg/kg	Arsenic	0.028 mg/kg
Cadmium	0.018 mg/kg	Copper	15.0 mg/kg
Lead	0.026 mg/kg	Mercury	<0.01 mg/kg
Selenium	0.28 mg/kg	Tin	0.048 mg/kg

Bacteriological:

Salmonella	not detected /25g
Standard Plate Count	<300 cfu/g
Enterobacteriaceae	<10 cfu/g
B. cereus	<100 cfu/g
Coagulase Positive Staph	<100 cfu/g
E.Coli	<3.0 MPN/g
Coliforms	3.6 MPN/g
Staphylococcus aureus	<10 cfu/g

Microbiological result*:

The results of the microbiological analysis are provided above. These results apply to powder which was produced under food grade conditions as described here. The results show that pathogens of potential concern were not detected and results for Enterobacteriaceae and total counts were satisfactory; therefore indicate that the powder as tested is suitable for human consumption (*confirmed and approved by the Food Risk Assessment Team, CSIRO Food & Nutrition Flagship).

Comments:

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(f) Product specification of the placebo powder

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Private Bag 16, Werribee VIC 3030, Australia
T +61 3 9731 3200 • ABN 41 687 119 230

Product Specification

Product:	Placebo																																																																
Date of Manufacture:	29 March 2015																																																																
Description:	Placebo powder is a freeze dried powder processed from food grade maltodextrin with added Halal compliance beef-based flavouring and colouring. This product appears as a brownish powder.																																																																
Process:	Maltodextrin powder is added with water, beef flavouring and colouring, then blended and frozen. The frozen placebo is freeze dried, milled, sieved and blended to meet specifications.																																																																
Specifications:	<u>Proximates:</u> <table style="width: 100%; border-collapse: collapse;"> <tr><td style="width: 60%;">Moisture</td><td style="width: 20%;"></td><td style="width: 20%; text-align: right;">2.6%</td></tr> <tr><td>Fat</td><td></td><td style="text-align: right;"><0.2%</td></tr> <tr><td>Protein</td><td></td><td style="text-align: right;">0.3%</td></tr> <tr><td>Ash</td><td></td><td style="text-align: right;"><0.1%</td></tr> <tr><td>Carbohydrate</td><td></td><td style="text-align: right;">97%</td></tr> </table> <u>Vitamin:</u> <table style="width: 100%; border-collapse: collapse;"> <tr><td style="width: 60%;">Vitamin A</td><td style="width: 20%;"></td><td style="width: 20%; text-align: right;"><5 µg/100g</td></tr> </table> <u>Trace Elements:</u> <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 33%;">Iron</td> <td style="width: 17%; text-align: right;"><2 mg/kg</td> <td style="width: 33%;">Zinc</td> <td style="width: 17%; text-align: right;">0.265 mg/kg</td> </tr> <tr> <td>Antimony</td> <td style="text-align: right;"><0.01 mg/kg</td> <td>Arsenic</td> <td style="text-align: right;"><0.01 mg/kg</td> </tr> <tr> <td>Cadmium</td> <td style="text-align: right;"><0.01 mg/kg</td> <td>Copper</td> <td style="text-align: right;">0.089 mg/kg</td> </tr> <tr> <td>Lead</td> <td style="text-align: right;">0.017 mg/kg</td> <td>Mercury</td> <td style="text-align: right;"><0.01 mg/kg</td> </tr> <tr> <td>Selenium</td> <td style="text-align: right;"><0.01 mg/kg</td> <td>Tin</td> <td style="text-align: right;"><0.01 mg/kg</td> </tr> </table> <u>Bacteriological:</u> <table style="width: 100%; border-collapse: collapse;"> <tr><td style="width: 60%;">Salmonella</td><td style="width: 20%;"></td><td style="width: 20%; text-align: right;">not detected /25g</td></tr> <tr><td>Standard Plate Count</td><td></td><td style="text-align: right;"><40 cfu/g</td></tr> <tr><td>Enterobacteriaceae</td><td></td><td style="text-align: right;"><10 cfu/g</td></tr> <tr><td>B. cereus</td><td></td><td style="text-align: right;"><100 cfu/g</td></tr> <tr><td>Coagulase Positive Staph</td><td></td><td style="text-align: right;"><100 cfu/g</td></tr> <tr><td>E.Coli</td><td></td><td style="text-align: right;"><3.0 MPN/g</td></tr> <tr><td>Coliforms</td><td></td><td style="text-align: right;"><3.0 MPN/g</td></tr> <tr><td>Staphylococcus aureus</td><td></td><td style="text-align: right;"><10 cfu/g</td></tr> </table>			Moisture		2.6%	Fat		<0.2%	Protein		0.3%	Ash		<0.1%	Carbohydrate		97%	Vitamin A		<5 µg/100g	Iron	<2 mg/kg	Zinc	0.265 mg/kg	Antimony	<0.01 mg/kg	Arsenic	<0.01 mg/kg	Cadmium	<0.01 mg/kg	Copper	0.089 mg/kg	Lead	0.017 mg/kg	Mercury	<0.01 mg/kg	Selenium	<0.01 mg/kg	Tin	<0.01 mg/kg	Salmonella		not detected /25g	Standard Plate Count		<40 cfu/g	Enterobacteriaceae		<10 cfu/g	B. cereus		<100 cfu/g	Coagulase Positive Staph		<100 cfu/g	E.Coli		<3.0 MPN/g	Coliforms		<3.0 MPN/g	Staphylococcus aureus		<10 cfu/g
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1 of 1

(g) Letter of importation from Indonesian collaborators



KEMENTERIAN PENDIDIKAN & KEBUDAYAAN
FAKULTAS KEDOKTERAN UNIVERSITAS PADJADJARAN
DEPARTEMEN ILMU GIZI MEDIK
Jl. Prof. Eijkman No. 38 Bandung Telp. (022) 2037234 Fax. (022) 2038305

Nomor : 079/UN6.C.1.1.10/TU/2015
Hal :

25 Mei 2015

To Whom It May Concern:

This is a statement of declaration that the importation of dried bovine product samples from Commonwealth Scientific and Industrial Research Organisation (CSIRO) in Australia into Indonesia is solely for non-commercial, research purposes.

Universitas Padjadjaran in Bandung, Indonesia (Dr Gaga Irawan Nugraha) is collaborating with the University of Otago, New Zealand (Dr Lisa Houghton) and CSIRO, Australia to investigate food-based strategies for preventing micronutrient deficiencies, in particular iron and zinc deficiency, which is a major concern in Indonesian infants.

Meat products are a rich source of micronutrients required for normal growth and development, including protein, iron and zinc and inadequate intakes of these nutrients can lead to stunting. There is good evidence that consumption of meat products, including beef meat and beef liver, between 6 and 12 months of age are important for preventing iron and zinc deficiency and supporting optimal growth and development in young children.

Our research to date indicates that nearly 20% of infants in Sumedang, West Java are stunted and that many are not meeting the World Health Organization dietary recommendations' relating to intake of iron and zinc-rich foods such as meat, fish and poultry (see appendix). Since barriers to consumption include food safety concerns and cost, we are exploring ways in which to improve access to meat products in a safe and economical way.

To that end, we have sought expertise from CSIRO in Australia, with financial support from Meat & Livestock Australia, to develop suitable dried meat products according to specifications required for food safety, eating and nutritional quality as well as being Halal certified.

Four prototypes have been developed, including a beef liver powder, beef meat powder, beef meat and liver powder blend, and a placebo powder made from starch (maltodextrin), beef flavouring and brown colouring. Each of these products will be packaged in sachets containing 15-20g.

The next phase of our research is to test the suitability of these prototype products in the home. To that end, a 2 week study involving 97 mother-infant pairs in Sumedang is planned. Mothers will add each of the products to the child's meal e.g. rice porridge and report on acceptance of the product by both the child and mother.

The results will inform the next phase of our research which will determine whether these products have the potential to correct micronutrient deficiencies found to be prevalent in this community.

The findings of this research will help to improve the lives of Indonesian children and achieve economic savings by reducing disease burden, improving school results and ultimately, adult work productivity. In addition, the collaboration supports sharing of expertise which is essential for building academic capacity at the Universitas Padjadjaran.

Sincerely yours,

Dr. Gaga Irawan Nugraha, dr., SpGK.M.Gizi
NIP. 19740305200012 1 002